

## Qualitative and Quantitative Analysis of Chocolates: An Undergraduate Science Experiment

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### Abstract:

The qualitative tests performed on different chocolate samples were for the following nutrients: proteins, fats and reducing sugars. A reducing sugar is a sugar that acts as a reducing agent, specifically, a type of carbohydrate that contains a free aldehyde or ketone group. Reducing sugars are important in food items because they can react with other parts of the food, like amino acids, to change the colour or taste of the food. The browning of food occurs due to the Maillard reaction, a process that occurs when a reducing sugar reacts with an amine group. This reaction usually occurs when food is heated or left at room temperature for a long period of time. The Maillard reaction adds to the flavour and aroma of many food items, such as coffee, chocolate and baked breads.

The minerals for which analysis was carried out were calcium, magnesium and iron. Quantitative analysis was performed in the chocolate samples for glucose, total reducing sugars and total fats. Glucose, physiologically known as blood sugar, is essential for brain function and physical energy.

**Key Words** : Qualitative, Quantitative, Chocolate, Coco, *Theobroma cacao*

### 1. Introduction :

A blend of cocoa powder, cocoa butter, and natural or synthetic sweeteners, chocolate can be solid or liquid. (Ewens et al, 2021) Modern chocolates may additionally contain milk solids, moderators, preservatives, and occasionally nuts. These can take the shape of several modern-day categories of syrups, bars, powders, and creams.

Since ancient times, people have enjoyed eating chocolate. Not only with ice cream and flavour-infused milk, but also with medication, chocolate syrup's lengthy history actually began. Throughout the most of its early history, chocolate was drunk as a bitter drink produced from fermented, roasted, and ground beans in its site of origin, Mesoamerica, or the region of North America that encompasses modern-day Mexico and nations to its south. *Theobroma cacao*, which literally translates to "food for the gods," is the cacao tree from whose beans the main ingredient was taken. Nowadays, the name "cacao" is used to describe the plant or its raw

beans; "cocoa" is the powdered extract of the cacao plant, and "chocolate" is anything prepared from the beans. (Watson, T., 2013)

It was frequently given for adults with wasting illness because the increased calories helped patients gain weight and the caffeine-like ingredients energised them. It was also thought to have aphrodisiac properties. There is proof that cultures living in Mexico made chocolate as early as 1900 BCE. In exchange for cacao beans, which they thought were a gift from Quetzalcoatl, the Aztec deity of wisdom, the Aztecs traded with the Mayans. As they were aware that cacao trees couldn't be produced in the heart of their civilisation, the Aztecs, who eventually overtook the Mayans as Mesoamerica's leading culture, continued to rely on them for cacao seeds. In fact, people used the seeds as money since they were thought to be so precious.(food.ndtv.com 2017)

After its arrival in Europe in the sixteenth century, sugars were added to it, which only resulted in its increased popularity. As the industrial revolution ushered in machinery that took over the time- intensive process of turning cacao to cocoa, the price of manufacturing dropped, the price of sugar dropped, and then we had chocolate bars. In the twentieth century, chocolates were considered essential ration for the US soldiers. From being considered a divine gift and a form of The Aztecs even traded cacao as currency. After its arrival in Europe in the sixteenth century, sugars were added to it, which only resulted in its increased popularity. As the industrial revolution ushered in machinery that took over the time-intensive process of turning cacao to cocoa, the price of manufacturing dropped, the price of sugar dropped, and then we had chocolate bars. In the twentieth century, chocolates were considered essential ration for the US soldiers.

There are several types of chocolate, classified according to the proportion of cocoa used in a particular formulation. Dark and bitter chocolates usually have high cocoa percentages ranging from 70% to 100%, while milk chocolates have an intermediate range (30%-45%, or even less) and white chocolates have none.

With their growing popularity among people of all age groups, especially among young children and teens, the nutritional value of chocolates becomes a matter of concern. High fat and sugar content is a major nutritional disadvantage of chocolates. This research was conducted to check for the presence of certain nutrients in popular brands of different categories of chocolates and to compare the amount of reducing sugars and fats in them.

### **1.1 Categories of Chocolates**

Chocolate is a sweet treat that is made from cocoa solids, cocoa butter, sugar, and sometimes milk or other ingredients. (Apshara, S. E. , Hubballi, N. V., 2013)The specific composition of chocolate can vary depending on the type and brand, but here are some common components:

**Cocoa solids:** These are the ground up pieces of roasted cocoa beans, and they are the primary ingredient in chocolate. Cocoa solids contain many different compounds, including flavonoids, theobromine, and caffeine, which give chocolate its characteristic flavor and aroma.

**Cocoa butter:** This is the fat that is extracted from cocoa beans during the chocolate-making process. Cocoa butter gives chocolate its smooth, creamy texture and helps to bind the other ingredients together.

**Sugar:** Most chocolate contains sugar to sweeten the flavor. The amount of sugar can vary depending on the type of chocolate, with dark chocolate typically containing less sugar than milk chocolate or white chocolate.

**Milk:** Some types of chocolate, such as milk chocolate and white chocolate, contain milk or milk powder. This adds a creamy flavor and texture to the chocolate.

**Emulsifiers:** These are added to help bind the ingredients together and create a smooth texture. Common emulsifiers include soy lecithin and polyglycerol polyricinoleate (PGPR).

**Other ingredients:** Depending on the brand and type of chocolate, other ingredients may be added, such as nuts, fruits, spices, or flavorings.

Overall, the composition of chocolate can vary widely depending on the type and brand, but cocoa solids, cocoa butter, sugar, and milk are the most common components.

There are several types of chocolate, each with a different composition and flavor profile. Here are some of the most common types of chocolate:

**Milk chocolate:** Milk chocolate is made with cocoa solids, cocoa butter, sugar, and milk powder or condensed milk. It has a creamy, sweet flavor and is the most popular type of chocolate in many countries.

**Dark chocolate:** Dark chocolate is made with a higher percentage of cocoa solids and less sugar than milk chocolate. It has a more intense, slightly bitter flavor and is often enjoyed by people who prefer a less sweet chocolate.

**White chocolate:** White chocolate is made with cocoa butter, sugar, and milk powder, but does not contain any cocoa solids. It has a creamy, sweet flavor and a smooth texture.

**Semi-sweet or bittersweet chocolate:** These types of chocolate are similar to dark chocolate, but have an even higher percentage of cocoa solids and less sugar. They have a more intense, slightly bitter flavor and are often used in baking.

**Couverture chocolate:** Couverture chocolate is a high-quality chocolate that is used by chocolatiers and pastry chefs. It has a higher percentage of cocoa butter, which gives it a glossy finish and makes it easier to melt and temper.

**Ruby chocolate:** Ruby chocolate is a relatively new type of chocolate that is made from ruby cocoa beans. It has a unique pink color and a fruity, slightly sour flavor.

Vegan chocolate: Vegan chocolate is made without any animal products, such as milk powder or butter. It is often made with plant-based milk, such as soy or almond milk, and can be found in many different varieties, including dark, milk, and white chocolate.

These are just a few of the most common types of chocolate, but there are many others available, each with its own unique flavor and texture.

## 2. Materials and Method

Six types of chocolate (coded C1,C2,C3,C4,C5,C6 ) and one chocolate syrup (coded S1) were purchased randomly from local market and analysed on the same day. The qualitative tests performed on different chocolate samples were for the proteins, fats and reducing sugars. A reducing sugar is a sugar that acts as a reducing agent, specifically, a type of carbohydrate that contains a free aldehyde or ketone group. Reducing sugars are important in food items because they can react with other parts of the food, like amino acids, to change the color or taste of the food. The browning of food occurs due to the Maillard reaction, a process that occurs when a reducing sugar reacts with an amine group. This reaction usually occurs when food is heated or left at room temperature for a long period of time. The Maillard reaction adds to the flavor and aroma of many food items, such as coffee, chocolate and baked breads.

The minerals for which analysis was carried out were calcium, magnesium and iron. Quantitative analysis was performed in the chocolate samples for glucose, total reducing sugars and total fats. Glucose, physiologically known as blood sugar, is essential for brain function and physical energy.

The different categories of chocolates analysed were:-

- **White Chocolate – Nestlé’s Milkybar (C1)**  
White chocolate is a chocolate confection typically made from a blend of cocoa butter, milk solids, sugar, milk fat and a fatty emulsifier. White chocolate does not contain chocolate solids (cocoa powder), and is characterized by a pale ivory color. The white chocolate used for analysis here is Nestlé's Milky bar. According to the pack’s ingredients, it contains proteins, sugars, fats and sodium.
- **Milk chocolate – Cadbury’s Dairy Milk (C2)**  
It is a solid chocolate made with milk in the form of milk powder, liquid milk or condensed milk. Bars of fine milk chocolate typically have a cocoa content between 30% and 45%, while less-expensive products may have considerably less. The chocolate analyzed here was Cadbury’s Dairy Milk. It contains milk, sugar, cocoa mass, cocoa butter, vegetable fat and emulsifiers and has a minimum 26% cocoa.
- **Dark Sweetened Chocolate – Bournville Rich Cocoa (C3)**  
Dark sweetened chocolates have at least 35% pure chocolate with some small amount

of sugar added. The chocolate sample used for analysis here is Cadbury's Bournville Rich Cocoa, with 50% cocoa content.

- **Bitter Chocolate – Amul Bitter 75% Cocoa Content (C4)**

Bitter chocolate is pure chocolate with very little or no added sugars. It has a higher percentage of cocoa with all fat content coming from cocoa butter instead of milk. The chocolate used for analysis is Amul Bitter Chocolate with 75% cocoa content.

- **Energy Bar – Patanjali Energy Bar (C5)**

Energy Bars are made to provide instant energy. They provide a highly nutritious, quite easily digestible and high carbohydrate based snack. The chocolate analyzed here is Patanjali Energy Bar.

- **Caramel Chocolate – Cadbury's 5 Star (C6)**

Caramel chocolates are generally made from butter, sugar, corn syrup, milk and cocoa. The chocolate contains caramel and nougat mix covered with smooth milk chocolate. The chocolate used for analysis here is Cadbury's 5 Star, which is sold in a golden wrapper decorated with stars.

- **Chocolate Syrup – Hershey's Chocolate Syrup (S1)**

Chocolate syrup usually consists of unsweetened cocoa powder, corn syrup or cane sugar, water and other flavorings.

The chocolate syrup analyzed here is Hershey's Chocolate Syrup. Hershey's Chocolate Syrup is low in fat and even has a bit of dietary fiber, including high fructose corn syrup.

## 2.1 Qualitative Analysis

### 2.1.1 Qualitative Analysis Test for Proteins

#### Chemicals Required

Chocolate solution: A small piece of solid chocolate is dissolved in warm water and the mixture is filtered. The filtrate so obtained is used to carry out the tests. NaOH pellets, Fehling's solution A, conc. HNO<sub>3</sub>, 40% NaOH solution

The following tests were performed:

#### i. Biuret Test

The Biuret test for proteins positively identifies the presence of proteins in solution with a deep violet color. Biuret, (H<sub>2</sub>NCONHCONH<sub>2</sub>), reacts with copper (II) ions in an alkaline solution to form a deep violet complex. The peptide linkages in proteins resemble those in Biuret and also form deep violet complexes with copper (II) ions in alkaline solution.

#### Procedure

5 mL of chocolate solution is taken in a test tube. A pellet of sodium hydroxide is added, followed by 1-2 drops of copper sulphate solution. Appearance of violet colour implies that protein is present.

## **ii. Xanthoproteic Test**

Xanthoproteic test is used to detect amino acids containing an aromatic nucleus (tyrosine, tryptophan and phenylalanine) in a protein solution which gives yellow colour nitro derivatives on heating with conc.  $\text{HNO}_3$ . The aromatic benzene ring undergoes nitration to give yellow coloured product. Phenylalanine gives negative or weakly positive reaction though this amino acid contains aromatic nucleus because it is difficult to nitrate under normal condition. On adding alkali to these nitro derivative salts, the colour changes from yellow to orange.

### **Procedure**

To a small amount of chocolate solution in a test tube, 2-3 drops of conc.  $\text{HNO}_3$  are added and the test tube is heated and then cooled under tap water. A yellow solution is obtained. To the solution, 2 mL of 40% NaOH is added – orange coloration – amino acids containing aromatic nucleus present.

### **2.1.2 Qualitative Analysis Test for Fats**

#### **Chemicals Required**

For liquid sample: ethanol

The tests were performed as follows: For solid sample

A small sample of chocolate is taken on a piece of filter paper. The paper is folded and unfolded to crush the sample. Appearance of translucent spot observed around the sample which indicates the presence of fats.

For liquid sample

A small amount of liquid chocolate is taken in a test tube and, ethanol is added. The mixture is shaken and filtered. Subsequently, water is added. Formation of an oily layer observed at the top of the water layer indicates the presence of fats.

### **2.1.3 Qualitative Analysis Test for Reducing Sugars**

#### **Chemicals Required**

Fehling's solution A  
Fehling's solution B  
Concentrated sulfuric acid

Tollen's reagent

Molisch's reagent: Prepared by dissolving a pinch of  $\alpha$ -naphthol in a small amount of ethanol.

The tests were performed as follows:

### **i. Molisch's Test**

Molisch's test is a general test for the identification of all carbohydrates and glycoprotein. In this test, conc. sulfuric acid is added to the solution which hydrolyses all the glycosidic linkages to form furfural and its derivatives, which are very reactive and condense with alpha-naphthol to give a purple or violet coloured product.

#### **Procedure**

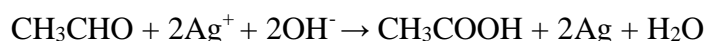
To 5 mL of chocolate sample in a test tube, 1 mL water and a few drops of freshly prepared Molisch's Reagent is added, followed by addition of conc.  $H_2SO_4$  along the sides of the test tube. Formation of a reddish purple ring at the interface indicates the presence of reducing sugars.

### **ii. Tollen's Test**

It is also known as Silver Mirror test. It is used for identification of carbohydrates containing alpha hydrogen. Tollen's reagent oxidizes an aldehyde into the corresponding carboxylic acid. This reaction is accomplished by the reduction of silver ions in Tollen's reagent into metallic silver, which, if the test is carried out in a clean glass test tube forms a mirror on the test tube.

#### **Procedure**

To 2 mL of freshly prepared Tollen's Reagent in a boiling tube, a pinch drop of chocolate solution is added and heated on a water bath. Formation of a silver mirror on the inner walls of the test tube indicates the presence of reducing sugars.

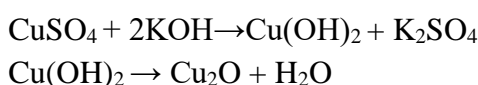


### **iii. Fehling's Test**

Fehling's test is a specific test for the identification of reducing sugars. When Fehling's reagents react with reducing sugars it reduces into brownish red precipitates. Oxidation of carbonyl group takes place which give aldonic acid. Formation of brownish red precipitates indicate the presence of reducing sugars in the sample.

### Procedure

Equal amounts of Fehling's A and Fehling's B solutions are mixed in a boiling tube., A pinch of chocolate sample is added and heated on a water bath. Formation of a reddish- brown precipitate of  $\text{Cu}_2\text{O}$  indicates the presence of reducing sugars.



## 2.1.4 Qualitative Analysis Test for Calcium

### Chemicals Required

Saturated ammonium chloride solution (A pinch of solid ammonium chloride was added to minimum amount of deionized water), 1:1 ammonia solution, saturated ammonium carbonate solution, conc. HCl

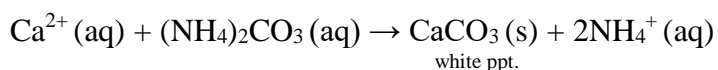
The following tests were performed:

### Precipitation test

The group 5 cations ( $\text{Ca}^{2+}$ ,  $\text{Sr}^{2+}$ ,  $\text{Ba}^{2+}$ ) are precipitated as their carbonates by addition of  $(\text{NH}_4)_2\text{CO}_3$  solution in presence of  $\text{NH}_4\text{Cl}$  and  $\text{NH}_4\text{OH}$ .

### Procedure

A small amount of chocolate solution is taken in a test tube. A solution of  $\text{NH}_4\text{Cl}$  is added, followed by ammonia solution and  $(\text{NH}_4)_2\text{CO}_3$  solution. Formation of a white precipitate indicates the presence of calcium.



### Flame Test

When metal salts are heated strongly in a flame, they are thermally ionized. The cation so formed absorbs heat energy and the valence electrons get promoted to higher energy level.



When the electrons drop back to the ground state, they release energy in the form of light which corresponds to a characteristic color. Since different metal ions emit light energy of different wavelengths depending on their energy levels, the color imparted by various salts is different. Calcium ions impart a brick-red color to the flame.

### Procedure

A small amount of solid chocolate sample is placed on a watch glass and made into a paste with a drop of conc. HCl. The paste is taken on the tip of a glass rod and introduced into the blue flame of a Bunsen burner. Observation of a brick red flame indicates the presence of Ca<sup>2+</sup>.

### 2.1.5 Qualitative Analysis Test for Magnesium

#### Chemicals Required

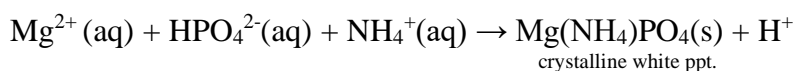
Saturated ammonium chloride, 1:1 ammonia solution, saturated ammonium phosphate

The test was performed as follows:

**Diphosphate Hydrogen Sulphate Test:** Magnesium gives white crystalline ppt. of magnesium ammonium phosphate, when excess of NH<sub>4</sub>Cl and NH<sub>4</sub>OH are added followed by addition of disodium hydrogen phosphate.

#### Procedure

To a small amount of chocolate sample in a test tube, NH<sub>4</sub>Cl solution and ammonia solution are added, followed by a saturated solution of ammonium phosphate. Formation of a white precipitate indicates the presence of Mg<sup>2+</sup>.



### 2.1.6 Qualitative Analysis Test for Iron

#### Chemicals Required

Ammonia solution, ammonium chloride

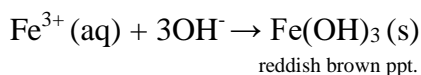
The following test was performed:

Fe<sup>3+</sup>, a Group 3 cation, may be precipitated as its hydroxide. The group reagent is NH<sub>3</sub> solution in presence of NH<sub>4</sub>Cl.



The presence of NH<sub>4</sub><sup>+</sup> in the second step suppresses the ionization of NH<sub>3</sub> in the first step and the equilibrium shifts backwards. The concentration of OH<sup>-</sup> decreases but this low

concentration is sufficient to cause precipitation of a Group 3 cation such as iron without allowing manganese or magnesium, if present, to precipitate.



## Procedure

A small amount of chocolate solution is taken in a test tube. Some ammonium chloride is added, followed by ammonia solution. Formation of a brown precipitate indicates the presence of iron.

## 2.2 Quantitative Analysis

### 2.2.1 Estimation of Reducing Sugars

**The Lane-Eynon method:** Lane and Eynon method ([egyankosh.ac.in](http://egyankosh.ac.in)) is based on the principle of reduction of Fehling's solution by reducing sugars. Fehling's solution is a mixture of copper sulfate and alkaline Rochelle salt (sodium potassium tartarate). Rochelle salt complexes with the cupric hydroxide formed in alkaline solution and prevent it from precipitation. Reducing sugars reduces the complexed cupric hydroxide to red, insoluble cuprous oxide under the experimental conditions. An oxidation-reduction indicator, usually methylene blue, detects the end point of the reaction.

Reducing sugars decolorize the methylene blue by reducing it to its leuco form. The active moiety is the sugar's aldehyde group.

Once all the copper sulfate in solution has reacted, any further addition of reducing sugars causes the indicator to change from blue to white. The volume of sugar solution required to reach the end point is recorded. The reaction is non-stoichiometric, which means that it is necessary to prepare a calibration curve by carrying out the experiment with a series of standard solutions of known carbohydrate concentration.

However, this method depends on the precise reaction times, temperatures and reagent concentrations used and so these parameters must be carefully controlled. It cannot distinguish between different types of reducing sugar, and cannot directly determine the concentration of non-reducing sugars. Also, it is susceptible to interference from other types of molecules that act as reducing agents.

### Apparatus Required

Chemical balance, Heating Mantle, 250 mL Volumetric Flask, 25 mL Burette, 100 mL Conical Flask, China Dish, small funnel, Whatman filter paper, Pumice stones

## Chemicals Required

Fehling's Solution A, Fehling's Solution B, Sucrose, 20% neutral lead acetate (5 g in 25 mL), 10% potassium oxalate solution (1 g in 10 mL), Methylene Blue Indicator

## Procedure

- i. Standardization of the Fehling's Solution with standard sucrose solution: Accurately weighed 4.75g of AR grade Sucrose. Transferred to a 500 mL volumetric flask with 50 mL distilled water. Added 5 mL conc. HCl and allowed to stand for 24 hours. Neutralized the solution with NaOH using phenolphthalein as indicator and made up to volume. Mixed well and transferred 25 mL to a 100 mL volumetric flask and made up to volume with distilled water. Transferred to the burette and titrated against Fehling's solution as described for sample.

## Preparation of chocolate sample

Weighed accurately 5 g sample of chocolate and transferred to 250 mL volumetric flask. Added about 50 mL warm water and neutralized with NaOH solution to phenolphthalein end point. Added 10 mL neutral 20% lead acetate solution, shook and let stand for 10 min. Added 10% potassium oxalate solution in small amounts until there is no further precipitation. Made up to the required volume, mixed the solution well and filtered through a Whatman filter paper. Transferred the filtrate to a 25mL burette.

## Preliminary Titration

Pipetted out 5 mL each of Fehling's A and B solutions into a 250 mL conical flask and mixed thoroughly. Added about 10 mL water and a few pumice stones. Dispensed the sugar solution from the burette. Heated the solution to boiling. Added 3 drops of methylene blue indicator. Continued addition of the sugar solution drop wise until the blue colour disappeared to a brick red end point. Maintained a boiling period of 3 min. Noted down the titre value.

## Final Titration

Pipetted out 5 mL each of Fehling's A and B solutions into 250 mL conical flask. Added sample chocolate solution about 0.5 mL less than the titre value of preliminary titration. Heated the flask to boiling. Added 3 drops of methylene blue indicator. Completed the titration within one minute by adding 2-3 drops of sugar solution at a time, until the indicator is decolorized (formation of reddish brown precipitate) Noted down the titre value. Repeated the titration twice and took the average value.

### 2.2.2 Estimation of total fats

Crude fat is often synonymous with ether extract and generally refers to free lipids that can be extracted into less polar solvents such as petroleum ether or diethyl ether. The method used here involves solvent extraction and weighing of the lipid residue after solvent evaporation. For crude fat, petroleum ether is often the preferred solvent as it is relatively non-polar and extracts most non-polar components (triacylglycerols, sterols, tocopherols and similar compounds), but does poorly at extracting the polar lipids, such as glycolipids and phospholipids).

Petroleum ether (pet ether), also known as benzine, Painter's naphtha, petroleum naphtha, naphtha ASTM, petroleum spirits, X4 or Ligroin, is a commonly used solvent due to its relatively low cost compared to other organic solvents. It is less hygroscopic than diethyl ether, is less flammable than diethyl ether, and is more selective for hydrophobic lipids than diethyl ether. It also has a relatively low boiling point to allow low temperature evaporation and leave no residue.

The ether solution containing fat is separated from rest of the contents of the chocolate using centrifugation. Centrifugation is a technique used for the separation of particles from a solution according to their size, shape, density, viscosity of the medium and rotor speed. The particles are suspended in a liquid medium and placed in a centrifuge tube. The tube is then placed in a rotor and spun at a definite speed. It speeds up the sedimentation process, resulting in the separation into a supernatant containing dissolved fat and a solid residue. The ether containing dissolved fat is poured into a beaker and the ether is left to evaporate, leaving behind fat.

### **Apparatus Required**

Centrifugation tubes, clean, dry, pre-weighed 100mL beakers, Centrifuge

### **Chemicals Required**

Petroleum ether, chocolate sample 1g

### **Procedure**

- i. Transferred 1g of finely crushed chocolate sample into centrifugation tube.
- ii. Filled 3/4<sup>th</sup> of the centrifugation tube with petroleum ether and shook well.
- iii. Centrifuged the suspension of solid in solvent until a liquid layer containing dissolved fat separated out.
- iv. Decanted the supernatant liquid in a pre-weighed 100 mL beaker. Covered it with aluminium foil with holes punched in it and kept it overnight until the ether completely evaporated, leaving behind only fat.
- v. Weighed the beaker containing fat and determined the weight of fat in the sample.

### 3. Result and Discussion

All chocolate samples gave positive tests for proteins, fats and reducing sugars. Only Nestle's Milkybar gave positive test for calcium. No chocolate sample contains magnesium or iron. The results of the qualitative analysis are given in Table 1. The amounts of reducing sugars and fats in each chocolate determined quantitatively are listed in Table 2.

**Table 1: Results of Qualitative Analysis of Chocolates**

S. No.	Sample Code	Presence/Absence of Nutrients					
		Protein	Fats	Reducing Sugars	Calcium	Magnesium	Iron
1	C1	Present	Present	Present	Present	Absent	Absent
2	C2	Present	Present	Present	Absent	Absent	Absent
3	C3	Present	Present	Present	Absent	Absent	Absent
4	C4	Present	Present	Present	Absent	Absent	Absent
5	C5	Present	Present	Present	Absent	Absent	Absent
6	C6	Present	Present	Present	Absent	Absent	Absent
7	S1	Present	Present	Present	Absent	Absent	Absent

**Table 2: Percentage of Reducing Sugars and Mass of Fat/gram of different Chocolates**

S. No.	Sample Code	% Reducing Sugar	Mass of fat per gram of chocolate/g
1	C1	12.05%	0.3260
2	C2	7.96%	0.2838
3	C3	2.18%	0.3481
4	C4	7.85%	0.3694
5	C5	8.79%	0.1482
6	C6	20.12%	0.1652
7	S1	18.57%	0.0494

#### Comparison of Reducing Sugars and Fats in different categories of Chocolates

Among all the chocolates, C6 contains the highest percentage of reducing sugars (20.12%), followed by S1 (18.57%). C1 contains lesser amount of reducing sugars than them but contain more than that in C2 and C5. C4 has an even lesser percentage, while C3 has the least amount of sugars as shown in figure 1.

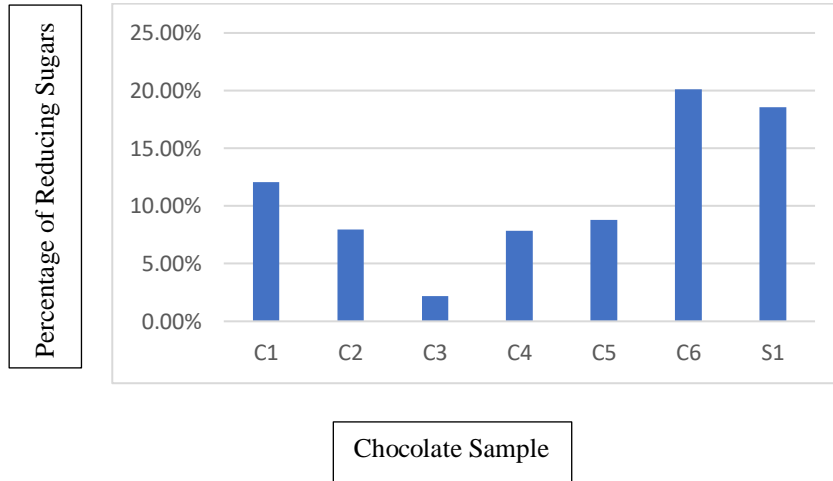


Figure 1 : comparison of percentage of reducing sugars in different chocolate samples

According to figure 2 C4 and C3 contain the highest amounts of fats (0.3694 g and 0.3481 g of fat per gram of respective chocolates). Since bitter and dark chocolates have higher contents of cocoa, their fat content generally comes from cocoa butter instead of milk. They are followed by C1 and C2 (0.326 g and 0.2838 g of fat per gram of respective chocolates). C5 and C6 have comparatively lesser amounts of fat, while S1 has the least.

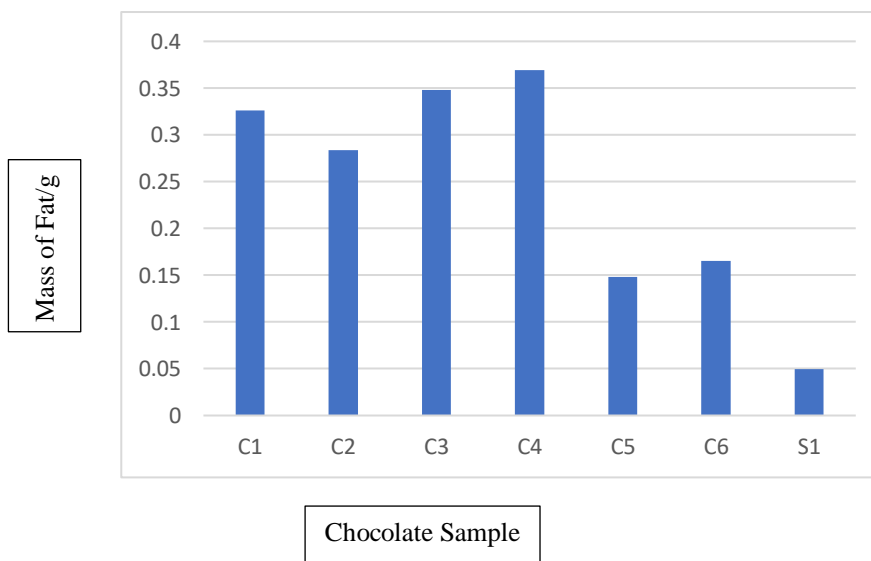


Figure 2 : comparison of fat content in different chocolate samples

#### **4. Conclusion**

Chocolates are rich sources of proteins, fats and reducing sugars. Caramel chocolates contain the highest amount of reducing sugars, while white and milk chocolates contain a lesser amount, dark and bitter chocolates containing the least. Most chocolates contain about 0.2-0.4 grams of fat per gram of chocolate. In this inquiry based learning activity determination of sugar and fat content of chocolates and qualitative analysis is done by utilising their knowledge of simple undergraduate chemical experiments. The learning activity is appropriate for a chemistry course in college or university since the topic is chosen from students' everyday lives and the outcomes of lessons can be applied to their daily lives.

#### **5. Declaration**

The manuscript has been prepared through contributions of all authors. All authors have given approval to the final version of the manuscript. All authors declare that they have no conflict of interest.

#### **6. Acknowledgements**

The present work has been carried out as part of an online summer research internship programme titled “Flavours of Research”, part of the D S Kothari Centre for Research and Innovation in Science Education, Miranda House, University of Delhi. Sujata Sengupta, Sharda Mahilkar Sonkar and Nutan Rani are thankful to the Principal, Miranda House, University of Delhi for her encouragement and motivation. Anand Sonkar is thankful to the Principal, Hansraj College, University of Delhi for her constant support and guidance.

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