Qualitative analysis of cyclamate in iced tea using varying BaCl$_2$ concentrations: A descriptive study in sekaran raya

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ABSTRACT

Cyclamate is one type of artificial sweetener that is widely used in beverages. However, excessive consumption can hurt health. This study aims to determine the presence of cyclamate content in iced tea products sold in the Sekaran Raya area around the campus of Semarang State University. The method used was descriptive research through qualitative analysis in the Integrated Science Laboratory with the precipitation technique. Three samples of iced tea drinks and one positive control sample (with cyclamate added) were analyzed by adding barium chloride (BaCl$_2$) reagent in an acidic atmosphere, with concentrations of 10% and 30%, respectively. The test results showed that one of the samples, iced tea sample RY, formed a white precipitate, indicating the presence of cyclamates, where 30% BaCl$_2$ produced more precipitate than 10% BaCl$_2$. This finding suggests that precipitation techniques with higher concentrations of BaCl$_2$ can improve the visibility of cyclamate detection. However, it should be noted that increasing the concentration of BaCl$_2$ is not always directly proportional to the identification quality because too high a concentration can cause interference and interfere with the clarity of the observation results.

1. INTRODUCTION

Artificial sweeteners are food additives that can cause a sweet taste in food and have no nutritional value (Permenkes, 1988), so they are often used as sugar substitutes in various foods or drinks. There are various types of artificial sweeteners (e.g., saccharin, cyclamate, aspartame, dulsin, and synthetic sorbitol) that arise due to several factors, including different levels of sweetness, stability under various conditions, and unique flavor profiles. For example, aspartame has a sugar-like sweetness, but is unstable at high temperatures, making it less suitable for baked products. On the other hand, cyclamate and saccharin have a higher level of sweetness, so they are only required in small amounts to achieve the desired sweetness. Each sweetener does have certain characteristics that make it suitable for specific uses. In addition, the diversity of artificial sweetener types is also influenced by consumer preferences, different safety regulations in each country, as well as innovations in research that are constantly evolving.

One of the frequently used artificial sweeteners is cyclamate, which is usually available in the form of Sodium cyclamate ($NaC_6H_{12}NO_3S$) or Calcium cyclamate ($CaC_6H_{12}NO_3S$) salts (Ukhdiiyah, 2018). Sodium cyclamate ($NaC_6H_{12}NO_3S$), or better known as cyclamate, has a sweetness level of about 30 to 50 times sweeter than sucrose (regular sugar) (Jamil et al., 2017; Lidiyawati, 2022). The advantage of cyclamate is that it is calorie-free, making it a safe choice for people with obesity and diabetes. The manufacturing process and circulation of cyclamate are always closely monitored and regulated by food regulatory agencies in various countries to ensure that its use does not exceed the set limits. The use of artificial sweeteners does need to be watched out for because, in excessive amounts, it will cause adverse side effects to health, such as tremors (nerve disorders), migraines, and headaches, decreased memory, confusion, hypertension (high blood pressure), and diarrhea (Melinda et al., 2022). This is especially felt by individuals who are sensitive to artificial sweetener compounds (Thahir & Masnar, 2021). The World Health Organization (WHO) states that there is a maximum limit of Sodium cyclamate ($NaC_6H_{12}NO_3S$) that can be consumed per day, or Acceptable Daily Intake (ADI), which is 11 mg/kg body weight. While in Indonesia, the use of artificial sweeteners is determined based on Permenkes 772/Menkes/Per/IX/88, for example, the maximum level of Sodium cyclamate ($NaC_6H_{12}NO_3S$) allowed in soft drinks is 3 g/kg (Marlina & Saa, 2019).

Drinks sold around boarding areas, especially by small traders or stalls, often lack supervision regarding the composition of the ingredients used. Therefore, testing the content of food additives, such as cyclamate, is important to ensure that the substance is within safe consumption limits to protect consumers. One method that can be used to qualitatively detect cyclamate content in beverage samples, such as iced tea, is the precipitation method. This method is commonly used in chemical analysis, both to separate and identify a compound (Khairunnisa, 2023). In the context of cyclamate analysis, the precipitation principle used is the reaction between cyclamate ions ($C_6H_{12}NSO_3^-$) and precipitating reagents that can form a characteristic precipitate. For example, barium chloride ($BaCl_2$) and silver nitrate ($AgNO_3$) are two commonly used reagents in this method due to their ability to react with cyclamate to form a water-insoluble precipitate of barium cyclamate ($BaC_6H_{12}NSO_3$) or silver cyclamate ($AgC_6H_{12}NSO_3$) (Mierza et al., 2023). The precipitate formed can be observed as an indication of the presence of cyclamate in the beverage sample. Although this method is qualitative, meaning that it does not provide quantitative information on the amount of the compound, it is still very useful as a first step for quick and simple food safety screening or surveillance in the field.

In the analysis of cyclamate content, precipitation occurs when the sample is added to barium chloride ($BaCl_2$) in an acidic state and heated in a water bath. The formation of a white precipitate indicates the presence of cyclamate compounds in the sample (Andini et. all, 2024). When barium

chloride ($BaCl_2$) reacts with cyclamate ions ($C_6H_{12}NSO_3^-$), a barium sulfate ($BaSO_4$) precipitate is formed, which generally appears as a white or yellowish-white solid that settles at the bottom of the test tube. This precipitate is insoluble in water, so it is easily isolated from the solution (Harahap et. all, 2023). If the sample does not contain cyclamate or if the level is very low, then the reaction with barium chloride ($BaCl_2$) does not produce a significant precipitate. Cyclamate itself is a type of artificial sweetener that is often used in food and beverage products, including iced tea drinks. Iced tea is a very popular drink and is often consumed by the public, including students around Semarang State University. Given the negative health impacts of excessive cyclamate consumption, it is important to identify the presence of this substance in beverage products in circulation. Therefore, this study aims to identify whether or not cyclamate is contained in samples of iced tea drinks sold by traders around Jalan Sekaran Raya, Universitas Negeri Semarang (UNNES), using the precipitation method.

2. METHODS

This study is descriptive research because it aims to describe the presence or absence of cyclamate content in samples of iced tea drinks sold by traders around Jalan Sekaran Raya, Semarang State University. A positive indication of the presence of cyclamate is indicated by the formation of a precipitate at the bottom of the test tube after the precipitation test. The laboratory test was conducted at the Integrated Science Laboratory, Faculty of Mathematics and Natural Sciences (FMIPA), Semarang State University. Three samples of iced tea drinks obtained from traders around Sekaran Raya, which is a dense student area, were chosen as the object of research because these drinks are commonly consumed by the UNNES academic community. This research is expected to provide an initial picture of the potential presence of artificial sweetener cyclamate in beverage products circulating in the campus environment.

A.   Tools and Materials

The tools used in the qualitative test of cyclamate by the precipitation method include 25 mL measuring cup (1 piece), 100 mL beaker glass (8 pieces), test tubes (10 pieces), tube rack (1 piece), 500 mL beaker glass (1 piece), drop pipette (4 pieces), funnel (2 pieces), filter paper (8 pieces), three legs (1 piece), spirtus (1 piece), test tube clamp (1 piece), wire gauze (1 piece), glass stirrer (1 piece), spatula (1 piece), and thermometer rod (1 piece). The materials used in the study were three samples of iced tea drinks (GB iced tea, OT iced tea, and RY iced tea) and one sample of iced tea as a positive control, 4 mL Hydrocloric Acid (HCl) 10%, 4 mL Barium Chloride ($BaCl_2$) 10%, 4 mL Sodium Nitrite ($NaNO_2$), cyclamate powder (2 spatulas) 10%, and distilled water.

B.   Research Procedure

The qualitative identification procedure of cyclamate by the precipitation method was carried out as follows.
1. Poured each iced tea sample into each beaker glass with code A for "GB" iced tea, code B for "OT" iced tea, code C for "RY" iced tea, and code D for the positive control sample.
2. In the sample that served as the positive control (code D), 2 spatula spoons of cyclamate powder were added to the beaker glass and stirred evenly.
3. From four beaker glasses, pour 3 mL of iced tea drink samples into test tubes.
4. Adding 3 mL of distilled water and 1 mL of hydrochloric acid (HCl) 10% to the iced tea sample. This acid is used to create the acidic conditions needed for the reaction.
5. After the sample is acidified, add 1 mL of barium chloride ($BaCl_2$) 10% solution to the sample. Stir gently with a glass stirrer, and let stand for 15-30 minutes.

6. Filter the mixture using filter paper and a funnel.
7. Added 1 mL of Sodium Nitrite (NaNO$_2$) 10% to the filtrate.
8. Heat the mixture using a three-legged stool and wire mesh (at an ambient temperature of 60-85°C) for 20-30 minutes. This heating accelerates the formation of the precipitate.
9. Observe and record the experimental results. If a white precipitate forms after the addition of BaCl$_2$ and heating, it can be concluded that the sample is positive for cyclamate.
10. Re-run steps 3-8 with barium chloride (BaCl$_2$) 30% concentration and coded E for "GB" iced tea, code F for "OT" iced tea, and code G for "RY" iced tea.

3. RESULTS AND DISCUSSIONS

The analytical method used to identify the cyclamate content in iced tea drink samples in this study is the precipitation method, which works on the principle of precipitate formation as a result of the reaction between the analyzed substance and certain reagents. The precipitate formed is a solid compound that is insoluble in the reaction medium, and its presence indicates that the precipitation reaction has occurred. The success of this precipitation process is greatly influenced by several factors, especially the solubility of compounds in solvents, temperature, and solution concentration. The lower the solubility of the compound formed, the greater the possibility of forming a stable precipitate that can be measured accurately. Therefore, precipitation should be carried out under well-controlled conditions to maximize precipitation and minimize possible interference from other compounds.

In this study, barium chloride (BaCl$_2$) reagent was used due to its ability to react with cyclamate ions (cyclamate anion C$_6$H$_{12}$NSO$_3^-$), forming a white precipitate of barium cyclamate (BaC$_6$H$_{12}$NSO$_3$), which is easily observed visually. To evaluate the sensitivity of the reaction to variations in reagent concentration, two barium chloride (BaCl$_2$) solutions of different concentrations, namely 10% and 30%, were applied to each sample. This method not only aims to detect the presence of cyclamate, but also to evaluate the intensity of the precipitate formed as an initial indicator of the relative content of cyclamate in the sample. The samples tested included samples of iced tea commonly consumed in the Sekaran Raya area, plus one control sample that had cyclamate added as a reference for comparison. Table 1 presents the qualitative analysis of the reaction time and precipitation observed in each sample treated with barium chloride (BaCl$_2$) 10%.

Table 1. Qualitative Analysis of Cyclamate in Iced Tea Samples using Barium Chloride (BaCl$_2$) 10% based on Reaction Time and Precipitation Observation

No	Sample	Addition of HCl (Hydrochloric acid)	Addition of 10% BaCl$_2$ (Barium chloride)	Time (seconds)	Sediment Observation	Conclusion
1	GB Iced Tea (A)	Dark brown (+++)	Clear brown (++)	813 seconds	There is no sediment (-)	Does not contain cyclamate (good)
2	OT Iced Tea (B)	Dark brown (++)	Clear brown (+)	768 seconds	There is no sediment (-)	Does not contain cyclamate (good)
3	RY Iced Tea (C)	Dark brown (+++)	Clear brown (++)	625 seconds	There is sediment (+)	Contains cyclamate (not good)
4	Positive Control (D)	Dark brown (+++)	Clear brown (+)	577 seconds	There is sediment (+++)	Contains cyclamate (not good)

Explanation of qualitative symbols:
(-) : No reaction or change (no sediment formed)
(+) : Weak reaction (slight color change or very little sediment)
(++) : Moderate reaction (noticeable color change or visible sediment)
(+++) : Strong reaction (clear color change or a large amount of sediment)

Note: The symbols (-), (+), (++), and (+++) represent the intensity of the observed reaction, indicating none, weak, moderate, and strong reactions, respectively, in terms of color change or sediment formation.

As a complement to the observation data displayed in tabular form, Figure 1 presents a visualization of the results of the precipitation test on four iced tea samples that have been treated with barium chloride ($BaCl_2$) reagent at a concentration of 10%. This visualization provides a clearer picture of the differences in the volume of precipitate formed in each sample. Variations in the amount of precipitate can be interpreted as an indication of the presence of barium cyclamate compounds, resulting from the reaction between barium ions (Ba^+) and cyclamate ions ($C_6H_{12}NSO_3^-$) in the sample. Of the four samples tested, the most striking precipitate was seen in the control sample (sample D) as well as one of the test samples (sample C). The presence of a fairly thick white precipitate in these two samples indicates a strong positive reaction between the cyclamate ions ($C_6H_{12}NSO_3^-$) and the barium chloride ($BaCl_2$) reagent, thus strengthening the suspicion that these two samples do contain significant amounts of cyclamate. Thus, this visualization not only reinforces the findings from the qualitative observations, but also provides a stronger basis in identifying which samples potentially violate the safe limits of food additive use as excessive consumption of sodium cyclamate has been shown to pose health risks such as metabolic disorders and increased oxidative stress (Hasan, et.al., 2023).
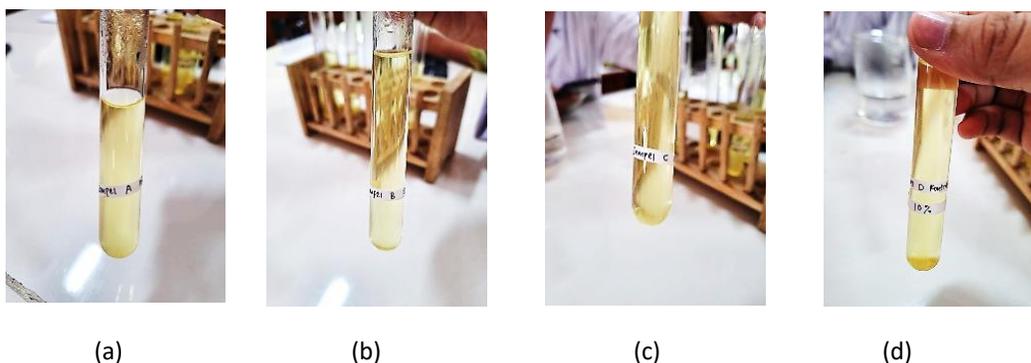

| (a) | (b) | (c) | (d) |

Figure 1. Visualization of the results of adding 10% $BaCl_2$ solution to each sample: (a) Sample A, (b) Sample B, (c) Sample C, and (d) Sample D

After initial testing using a 10% barium chloride ($BaCl_2$) solution, the study continued with the use of a higher reagent concentration, namely 30%. The purpose of this stage is to evaluate the extent to which increasing the reagent concentration affects the amount of precipitate formed. By increasing the reagent concentration, it is expected that the precipitation reaction will take place faster and produce a more clearly visible precipitate, thus facilitating the visual identification process. The iced tea sample, which in the initial test showed a positive possibility of containing cyclamate, was again analyzed using a 30% barium chloride ($BaCl_2$) solution. The results of the two reagent concentrations

were then compared to observe differences in the intensity and clarity of the precipitate, which can give further indication of the relative levels of cyclamate in each sample. A visual comparison between the precipitation reactions using 10% and 30% barium chloride ($BaCl_2$) for the same sample is presented in Figure 2, while Table 2 presents the qualitative analysis of the reaction time and precipitation observed in each sample treated with 30% barium chloride ($BaCl_2$).

Table 2. Qualitative Analysis of Cyclamate in Iced Tea Samples using Barium Chloride ($BaCl_2$) 30% based on Reaction Time and Precipitation Observation

No	Sample	Addition of HCl (Hydrochloric acid)	Addition of 10% BaCl₂ (Barium chloride)	Time (seconds)	Sediment Observation	Conclusion
1	GB Iced Tea (E)	Dark brown (+++)	Clear brown (++)	161 seconds	There is no sediment (-)	Does not contain cyclamate (good)
2	OT Iced Tea (F)	Dark brown (++)	Clear brown (+)	144 seconds	There is no sediment (-)	Does not contain cyclamate (good)
3	RY Iced Tea (G)	Dark brown (+++)	Clear brown (++)	132 seconds	There is sediment (++)	Contains cyclamate (not good)
4	Positive Control (H)	Dark brown (+++)	Clear brown (+)	120 seconds	There is sediment (+++)	Contains cyclamate (not good)

Explanation of qualitative symbols:
(-) : No reaction or change (no sediment formed)
(+) : Weak reaction (slight color change or very little sediment)
(++) : Moderate reaction (noticeable color change or visible sediment)
(+++) : Strong reaction (clear color change or a large amount of sediment)

Note: The symbols (-), (+), (++), and (+++) represent the intensity of the observed reaction, indicating none, weak, moderate, and strong reactions, respectively, in terms of color change or sediment formation.
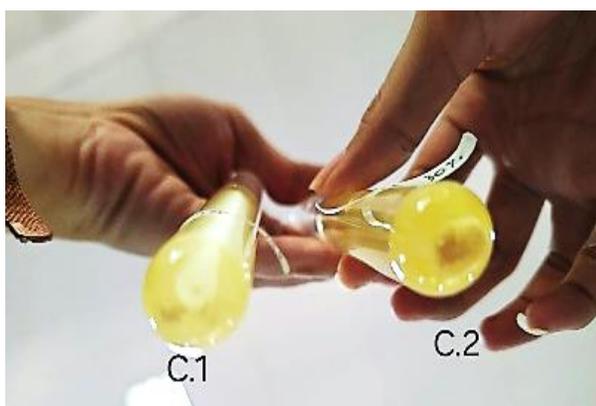

Figure 2. Sample C1 (Barium Chloride, $BaCl_2$ 10%) produced less precipitate than C2 (Barium Chloride, $BaCl_2$ 30%).

As explained earlier, the main principle in testing the cyclamate content in this study is the formation of a white precipitate of barium sulfate ($BaSO_4$). This precipitate is the result of the reaction between barium ions (Ba^{2+}) derived from barium chloride ($BaCl_2$) solution with sulfate ions (SO_4^{2-}) released from cyclamate compounds. Before the reaction takes place, the sample is first acidified with a 10% hydrochloric acid (HCl) solution of 1 mL. The purpose of this acidification is to convert cyclamate sodium salt ($C_6H_{11}NHSO_3Na$) into a free acid form, namely cyclamic acid ($C_6H_{12}NSO_3H$), which is more reactive towards barium ions (Ba^{2+}). The acidic conditions (low pH) produced by hydrochloric acid (HCl) allow the protonation process of cyclamate ions ($C_6H_{12}NSO_3^-$) to occur, so that the sulfate group in the cyclamic acid molecule becomes easier to interact with barium ions (Ba^{2+}) and accelerates precipitate formation (Ramadhani et al., 2018; Adriani & Aidil, 2021). In addition to hydrochloric acid (HCl), sodium nitrite ($NaNO_2$) solution is also added to the reaction process. Sodium nitrite ($NaNO_2$) plays an important role in helping to break the sulfate bond of the cyclamate compound, thus facilitating the formation of barium sulfate ($BaSO_4$) precipitates (Mierza et al., 2023). To ensure the reaction runs optimally, the solution mixture is shaken so that it is homogeneous and all components can react evenly.

The use of two concentrations of barium chloride ($BaCl_2$) solution, namely 10% and 30%, showed a significant effect on the amount and speed of formation of barium sulfate ($BaSO_4$) precipitate in cyclamate content testing. The higher concentration of the reagent, 30%, produced a larger amount of precipitate and a shorter time than the 10% barium chloride ($BaCl_2$) solution. This is due to the increased amount of barium ions (Ba^{2+}) in the solution, which directly increases the frequency of collisions between barium ions (Ba^{2+}) and cyclamate ions ($C_6H_{12}NSO_3^-$). More frequent collisions increase the chance of insoluble complexes forming, thus accelerating the precipitation process (Watung & Utama, 2023). These findings underscore the importance of reagent concentration as a crucial factor in determining the sensitivity, efficiency, and precision of a test method. In the precipitation reaction between barium ions (Ba^{2+}) and cyclamate ions ($C_6H_{12}NSO_3^-$), 1:1 stoichiometry applies, meaning that one mole of cyclamate ions ($C_6H_{12}NSO_3^-$) reacts with one mole of barium ions (Ba^{2+}) to produce one mole of barium sulfate ($BaSO_4$) precipitate. The precipitate formed is stable, insoluble in water, and easily separated from the liquid medium. These properties make barium sulfate ideal as a visual indicator in qualitative analysis (Andalia & Alfalah, 2021; Melinda et al., 2022).

Based on the test results of the iced tea drink samples, it was found that samples A, B, E, and F did not show the formation of precipitates, indicating that these samples did not contain cyclamate. Conversely, in samples C and D, as well as G and H, the formation of a white precipitate was observed, indicating a positive reaction for the presence of cyclamate. During the reagent addition process, the mixture was shaken to ensure even mixing, thereby ensuring homogeneity among the sample, reagent, and solvent so that the reaction could proceed optimally. Heating was also carried out to accelerate the chemical reaction rate. The heating was done using a water bath at a temperature between 60°C and 85°C for 20–30 minutes. During this process, a pungent odor emerged, which indicated the formation of nitrogen gas, signifying that the reaction was actively occurring. The temperature range used is categorized as moderate, deemed sufficient to increase the kinetic energy of the molecules without damaging the chemical structure of the compounds being analyzed (Mao et al., 2023; Melinda et al., 2022). This increase in kinetic energy plays a role in enhancing the likelihood of effective collisions between molecules, thereby accelerating the reaction between cyclamate ions ($C_6H_{12}NSO_3^-$) and the reagent. The visualization of the precipitation reaction observed in each sample can be seen further in Figure 3.
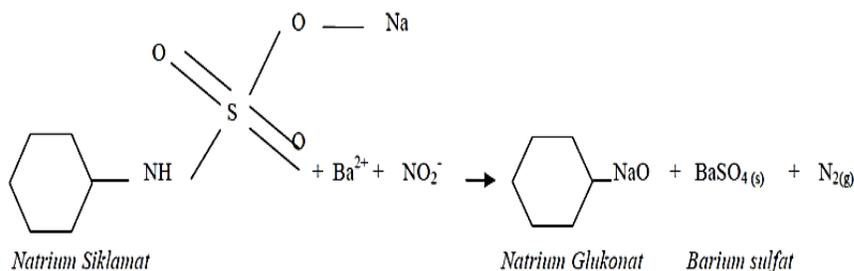
Figure 3. Reaction of formation of barium sulfate (BaSO₄) precipitate

Overall, the precipitation method has proven to be a simple, efficient, and sufficiently sensitive technique for detecting the presence of cyclamate in beverage samples. Its main advantage lies in its ability to produce a stable precipitate that is easily observable visually, without the need for complex or expensive laboratory equipment. This makes the method highly suitable for use in field conditions or laboratories with limited facilities. In addition, due to its relatively quick and easily applicable procedure, this method enables the early detection of artificial sweeteners such as cyclamate in a practical and economical way, offering added value in the context of quality control and food safety monitoring—particularly for beverage products that may contain food additives exceeding the permitted limits. In this regard, precipitation can serve as an effective preliminary screening step before conducting further quantitative analyses that require high-precision laboratory instruments, such as liquid chromatography or mass spectrophotometry. Thus, the precipitation method is not only technically relevant but also strategically important in supporting consumer protection efforts against the risks of exposure to harmful additives in everyday food and beverages.

4. CONCLUSIONS

Based on the results of the analysis and testing conducted, it can be concluded that the precipitation method using barium chloride (BaCl₂) reagent is effective for the qualitative analysis of the artificial sweetener cyclamate in iced tea beverage samples. The reaction between barium ions (Ba^{2+}) and sulfate ions (SO_4^{2-}) from the cyclamate compound produces a white barium sulfate (BaSO₄) precipitate that is visually observable, making it a clear indicator of the compound's presence. Among the three iced tea samples tested, only one sample (RY iced tea) showed the formation of a precipitate, indicating the presence of cyclamate in that product. Furthermore, testing with two different reagent concentrations revealed that a 30% barium chloride (BaCl₂) solution produced more and faster precipitate formation compared to the 10% concentration. These findings confirm that increasing the reagent concentration can enhance detection sensitivity, although this must be balanced against potential visual interference or side reactions that may occur at excessively high concentrations.

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