Chemical Preservation Assessment: Evaluating the efficacy and quantity of chemical preservatives like sodium benzoate.

[1]Roshini K.T, [2]Akshara Krishnan

[1]Assistant Professor
[1]Department of Chemistry,
[1]Carmel College, Mala, India

Abstract : Chemical preservatives play a crucial role in maintaining the safety and quality of food products, particularly in beverages like fruit juices where microbial spoilage is a significant concern. Sodium benzoate, a commonly used preservative, is known for its antimicrobial properties and widespread application in the food industry. In this study, we aimed to assess the efficacy and quantify the presence of sodium benzoate as a preservative in various fruit juice samples through rigorous chemical analysis**.** Fruit juice samples from different manufacturers were collected and subjected to analytical methods. Additionally, the pH of the samples was measured to understand its potential influence on the preservative's efficacy. Our results revealed significant variations in the concentration of sodium benzoate among the tested fruit juice samples. While some products exhibited levels within regulatory limits, others exceeded permissible thresholds, raising concerns about potential health implications and regulatory compliance.

***Index Terms* -** Chemical preservatives, Sodium benzoate, Efficacy, Permissible thresholds**.**

I. Introduction

Preservation of food and beverages is essential to maintain their safety, quality, and shelf life, particularly in the context of perishable products like fruit juices. Chemical preservatives, including sodium benzoate, are commonly utilized to inhibit microbial growth and delay spoilage, thus ensuring the longevity of food and beverage items. However, the effectiveness and safety of these preservatives warrant continuous assessment and scrutiny due to their widespread use and potential health implications. The soft drink industry is the leading consumer of benzoate as a preservative, primarily due to the prevalence of high fructose corn syrup in many carbonated beverages. Despite their inherent resistance to spoilage due to acidity and carbonation, soft drinks require preservatives to maintain stability during prolonged storage periods. Although sodium benzoate is generally recognized as safe (GRAS), short-term exposure to it can lead to irritation of the eyes, skin, and respiratory tract. Prolonged or repeated exposure may even cause skin sensitization, posing heightened risks for children who tend to consume larger quantities per kilogram of body weight, driven by their unique dietary patterns and preferences. Alarmingly, approximately 80% of children's exposure to benzoic acid and its salts stems from soft drinks and pre-packaged beverages.

Maintaining stringent cleanliness standards is paramount when employing sodium benzoate as a preservative. While preservatives like sodium benzoate play a crucial role in food preservation, they cannot substitute for proper hygiene in food processing. It's imperative to understand that sodium benzoate cannot salvage already spoiled products; its effectiveness hinges on the preservation of product integrity from the outset.

Sodium benzoate (E211) is a widely employed chemical preservative known for its antimicrobial properties and ability to inhibit the growth of bacteria, yeast, and mold in food and beverage products. It is commonly utilized in acidic foods and beverages, including fruit juices, soft drinks, and salad dressings, owing to its solubility and stability under acidic conditions. Despite its extensive use and regulatory approval in many countries, concerns have been raised regarding its safety, particularly when combined with certain additives or under specific storage conditions.

The efficacy of sodium benzoate as a preservative is influenced by various factors, including its concentration in the product, pH level, temperature, and presence of other additives. Additionally, regulatory authorities such as the Food and Drug Administration (FDA) and the European Food Safety Authority (EFSA) have established maximum allowable limits for sodium benzoate in food and beverage products to ensure consumer safety. However, adherence to these regulatory standards and the actual concentration of sodium benzoate in commercial products may vary, necessitating systematic evaluation and monitoring.

In this study, we aim to assess the efficacy and quantify the quantity of chemical preservatives, with a focus on sodium benzoate, in selected fruit juice samples. Through rigorous chemical analysis techniques, including high-performance liquid chromatography (HPLC) coupled with ultraviolet (UV) detection, we will determine the concentration of sodium benzoate in various commercially available fruit juices. Additionally, we will investigate the relationship between pH levels, preservative efficacy, and microbial stability to gain insights into the factors influencing preservation effectiveness. By conducting this chemical preservation assessment, we seek to contribute to the existing body of knowledge regarding the use of chemical preservatives in food and beverage products, particularly in the context of fruit juices. Our findings will not only provide valuable information for consumers, regulatory agencies, and food manufacturers but also contribute to enhancing the safety and quality of food products in the market.

II. Materials and Methods

2.1 Reagents and Chemicals: - Pure and analytical grade chemicals were used in all experiments

2.2 Mango juice samples: - Seven mango juice samples from different brands, all containing the approved class II preservative sodium benzoate, were thoughtfully selected and procured from local markets. Care was taken to ensure that the bottles were tightly sealed and devoid of any leaks. To maintain their freshness and integrity, the samples were refrigerated until they were ready for analysis. For the analytical process, precisely 10 grams of each mango juice sample was measured and transferred into separate beakers. Next, to alkalize the samples, 1ml of 10% NaOH solution was added to each beaker. Subsequently, 12 grams of sodium chloride were introduced into each sample. Thorough mixing ensued to ensure proper homogenization of the mixture. Afterward, the mixtures were allowed to stand undisturbed for several minutes to facilitate complete reaction and prepare them adequately for subsequent analysis.

2.3 Preliminary analysis to detect the presence of the preservative content

The samples underwent acidification with HCl and extraction with diethyl ether. Following this, the solvent was evaporated on a hot water bath to remove any remaining traces of solvent under a stream of air. The resulting residue was dissolved in a small amount of hot water. A few drops of 0.5% ferric chloride solution were then added to the solution. The formation of a salmon-colored precipitate of ferric benzoate served as an indication of the presence of benzoic acid in the samples.

2.4 Estimation of Sodium benzoate

To the mixture, one drop of phenolphthalein indicator was introduced, resulting in a noticeable color change. Subsequently, a few drops of dilute HCl were added until the color vanished, signifying neutralization. Excess 3ml of dilute HCl was then added to ensure complete acidification of the mixture. To this prepared solution, 25ml of chloroform was added, and the entire content was transferred into a 100ml separation funnel. It was allowed to stand for 20 minutes with periodic gentle shaking to facilitate the extraction process. Any emulsion formed during shaking was carefully broken by gently stirring with chloroform to avoid its formation. After sufficient extraction time, the lower chloroform layer, containing the target compound, was carefully transferred into a clean beaker. The solvent was then evaporated on a steam bath to concentrate the sample. Subsequently, 50ml of 50% ethanol solution was added to the evaporated residue. The resulting solution was then titrated with standardized NaOH solution, using phenolphthalein as an indicator. The NaOH solution had been previously standardized with oxalic acid to ensure accuracy. Finally, based on the volume of NaOH solution required for neutralization, the amount of sodium benzoate present in the given sample was calculated using appropriate stoichiometry and concentration factors.

1 ml of 0.05 M NaOH contains 0.0072g of sodium benzoate

% of sodium benzoate = (wt. of sodium benzoate / wt. of sample) x 100

2.4 Determination of pH

The pH of a solution is defined as the negative logarithm to the base 10 of the hydronium ion. Concentration in moles per litres The P^H meter was rinsed with deionised water and placed it in a beaker containing the buffer solution of P^H 7. Allowed the reading to stabilize. The sample fruit juice was taken in a clean 100 ml beaker and immersed the electrode into it. Allowed the reading to stabilize for about 2 minutes and read the display to obtain the P^H of the sample.

2.5 Determination of Total Dissolved Solids

Total dissolved solids present in the fruit juices was determined by the evaporation method. A clean 100 ml beaker was previously weighed on an electronic balance. 10 ml of different samples of fruit juices were taken in different beakers and heated to make it slurry. It is then kept in the oven for 3 hours at a temperature of 100 ^{0}C for drying. The weight of beakers along with dry samples were determined in an electronic balance. Subtracting the weight of empty beaker from this, we can calculate the amount of total dissolved solids. The percentage of TDS is then determined.

$$TDS = \frac{\text{Weight of beaker with the dry sample} - \text{Weight of empty beaker}}{\text{Weight of the liquid sample} - \text{weight of empty beaker}} \times 100$$

2.6 Nutritive Analysis of fruit juice samples

2.6.1. Test for Carbohydrate

Molisch's test: To small samples of synthetic fruit juices of different brands, 2 drops of 1% alcoholic α-naphthol solution was added. Add about 1 ml of Con. H_2SO_4 carefully along the sides of the test tube. A violet ring at the junction of the two layer shows the presence of carbohydrate.

Fehling's solution test: Small samples of synthetic fruit juices of different brands were taken in test tubes and few drops of Fehling A solution and Fehling B solution was added in equal amount. The test tube was heated in water bath for 10 minutes. Appearance of brown precipitate confirmed the presence of Glucose in synthetic fruit juices.

2.6.2. Test for Proteins

Xanthoproteic test: About 2 ml of concentrated aqueous solution of the compound add to about 0.75 ml of conc. HNO_3. A white crystalline precipitate indicates the presence of protein.

2.6.3 Test for starch: A little of the samples was added treated with few drops of iodine solution. Blue- black solution indicates the presence of starch.

2.6.4. Test for Sodium: The mixture is made up a paste with con. HCl on a watch glass. A little of paste is shown to the flame. A golden yellow flame indicates, presence of sodium.

2.6.5. Test for Potassium: Take little of the sample solution in the test tube and add 2ml of Picric acid shake for some time. Formations of yellow precipitate indicate the presence of potassium ion.

2.6.6. Test for Iron: Take a little of the sample in a test tube and add 1% ammonium thiocyanate solution. A blood red colouration observed indicates presence of Ferric iron

2.6.7. Test for Magnesium: A little of sample was treated with few drops of Magneson reagent followed by excess of NaOH solution. Blue precipitate indicated the presences of magnesium.

2.6.8 Test for Aluminium: To the drop of the sample solution add a drop of two molar sodium hydroxide, one drop of 1% aqueous solution of alizarin reagent and acetic acid in drops until violet disappears and one drop excess. A red precipitate observed indicates presence of Aluminium

2.6.9. Test for calcium: Two drops of sample solution acidified with acetic acid and three drops of saturated solution of Picrolinic acid in water are added. Presence of white crystals indicated presence of calcium.

III. Results and Discussion

The permissible limit of sodium benzoate as preservative in synthetic juices is 0.05-0.1%. The findings from the experimental observations (Table:1) indicate that all seven samples of synthetic fruit juices exceeded the permissible limit for sodium benzoate as a preservative, posing potential health risks due to the elevated levels of this additive. Among these samples, sample 6 exhibited the highest concentration of sodium benzoate (1.638%), while sample 5 had the lowest concentration (0.31%). This suggests that consumers should be cautious when consuming these synthetic fruit juices, as their sodium benzoate content exceeds the recommended safety limits. High levels of synthetic preservatives like sodium benzoate can contribute to various health concerns, including allergic reactions, hyperactivity, and potential carcinogenic risks associated with the formation of benzene under certain conditions. It's essential for manufacturers to ensure compliance with regulatory limits and for consumers to be vigilant about the ingredients in the products they consume. Choosing natural or minimally processed alternatives can help reduce exposure to lab-synthesized sodium benzoate and benzoic acid, when added to foods as preservatives, can indeed pose health risks when consumed in excessive amounts. While naturally occurring sodium benzoate found in fruits is generally harmless, the synthetic versions used as additives in processed foods and beverages can have adverse effects.

Table:1 Quantity of Sodium Benzoate in fruit juice samples

Sl.No.	Different brands of mango juices	% of Anhydrous Sodium Benzoate
1.	Sample 1	0.823
2.	Sample 2	1.085
3.	Sample 3	0.839
4.	Sample 4	1.5
5.	Sample 5	0.31
6	Sample 6	1.638
7.	Sample 7	1.253

The effectiveness of sodium benzoate as a preservative is influenced by the pH value of the product it's used in. Sodium benzoate is most effective as a preservative in acidic environments, with pH values below 4.5. In such environments, the ratio of undissociated (free) benzoic acid to ionized benzoic acid is higher, increasing its antimicrobial activity. Therefore, the pH value of the product is a critical factor in determining the efficacy of sodium benzoate as a preservative. Our analysis on P^H, reveals that among the given samples, samples 1, 2, and 3 have a pH of 3.9, while sample 7 has a pH of 2.8, making it the lowest pH value among the samples. This indicates that sample 7 is more acidic compared to samples 1, 2, and 3. Fruit juices are generally acidic. The low values for pH indicate this fact. pH values of fruit juices of different brands are given in Table 2 below. while acidic pH levels in foods and beverages are generally safe and can be part of a healthy diet, excessive consumption or prolonged exposure to highly acidic substances may have negative implications for dental, digestive, and overall health. Maintaining dietary balance and moderation is key to supporting optimal health and well-being.

Table:2 P^H Value of Fruit juice samples

Sl.No.	Juice Samples	P^H Values
1	Sample 1	3.9
2	Sample 2	3.9
3	Sample 3	3.9
4	Sample 4	3.7
5	Sample 5	3.4
6	Sample 6	3.1
7	Sample 7	2.8

Total dissolved solids (TDS) in fruit juices refer to the concentration of all inorganic and organic substances dissolved in the juice, including sugars, minerals, vitamins, acids, and other compounds. The TDS content can vary depending on factors such as the type of fruit, ripeness, processing methods, and added ingredients. High TDS levels can contribute to the taste, texture, and nutritional content of fruit juices. However, excessively high TDS levels may indicate contamination, poor quality, or excessive processing.

Among the different brands of fruit juices, that we selected, sample 7 contained the lowest solid content (9.4%) and sample 5 has the highest (23.4%).

Table: 3 Quantity of Total Dissolved Solids in fruit juice samples

Sl. No.	Fruit juices	% TDS
1	Sample 1	17.79
2	Sample 2	18.7
3	Sample 3	16.09
4	Sample 4	13.8
5	Sample 5	23.4
6	Sample 6	18.58
7	Sample 7	9.4

In general, fruit juices with moderate TDS levels are considered desirable as they provide flavor and nutritional benefits. However, it's essential to monitor TDS levels to ensure product quality and compliance with regulatory standards. Excessive TDS levels may affect the sensory properties of the juice, such as taste and clarity, and may also impact consumer acceptance.

Table: 4 Nutritive Analysis in fruit juice samples

Sl. No	Potassium	Sodium	Magnesium	Aluminium	Iron	Calcium	Starch	Proteins	Carbohydrates
Sample 1	✓	✓	✓	✓	×	×	✓	×	✓
Sample 2	✓	✓	✓	✓	×	×	✓	✓	✓
Sample 3	×	✓	✓	✓	×	×	✓	✓	✓
Sample 4	✓	×	✓	✓	×	×	✓	×	✓
Sample 5	✓	✓	✓	✓	×	×	✓	×	✓
Sample 6	×	✓	✓	✓	×	×	✓	×	✓
Sample 7	✓	✓	×	✓	×	×	✓	×	✓

Most of the selected fruit juice samples exhibited the presence of carbohydrates, proteins, starch, potassium, sodium, magnesium, and aluminium during their nutritive analysis.

IV. CONCLUSION

The determination of sodium benzoate content as a preservative in synthetic fruit juices through titration is a convenient, safe, and relatively quick method. Benzoic acid is isolated from a known quantity of sample by saturating it with sodium chloride, acidifying with dilute HCl, and extracting with chloroform. The extracted benzoic acid is then quantified through titration against standard alkali. The permissible limit of sodium benzoate in synthetic juices is 0.05-0.1%, but experimental findings revealed that all seven samples analysed exceeded this safe threshold. The pH values of the samples ranged from 3.1-3.9, indicating an acidic nature. The elevated acidity was attributed to the presence of mineral acids. Sodium benzoate's effectiveness as a preservative increase with decreasing pH due to a higher ratio of undissociated benzoic acid, which is the active antimicrobial agent. A comparative analysis of the percentage of total dissolved solids showed that sample 5 contained the highest concentration. Qualitative analysis detected the presence of carbohydrates, proteins, starch, potassium, sodium, magnesium, and aluminium in most of the samples.

REFERENCES

[1] Turkoglu, S. 2007. Genotoxicity of five food preservatives tested on root tips of Allium cepa L. Mutat Res., 626(1-2):4–14.

[2] Hong, H., Liang, X. and Liu, D.2009. Assessment of benzoic acid levels in milk in china. Food Control. 20(4):414–18.

[3] Oyewole, O.I., Dere, F.A. and Okoro, O.E. 2012 Sodium benzoate mediated hepatorenal toxicity in wistar rat: Modulatory effects of azadirachta indica (neem) leaf. Eur J Med Plant. 2(1):11.

[4] Lu, N. and Shen, M. 2006 Research on mutagenicity of sodium benzoate in born marrow cells. J Jilin Agri Univ. 28(4):466–68.

[5] Toth, B.1984. Lack of tumorigenicity of sodium benzoate in mice. Fundam Appl Toxicol. 4(3):494–96.

[6] Somya, K.V., Ravishankar, K., Basha, D.P. and Kiranmayi, G.V.N. 2011.Estimation of caffeine and sodiun benzoate in caffeine and sodium benzoate injection by isoabsorption method. Int J Pharm Chem Biol Sci. 1(1):26–31.

[7] Zhang, G and Ma, Y. 2013. Spectroscopic studies on the interaction of sodium benzoate, a food preservative, with calf thymus DNA. Food Chem. 141(1):41-47

[8] Zengin, N., Yüzbasıoglu, D., Ünal, F., Yilmaz, S. and Aksoy, H. 2011. The evaluation of the genotoxicity of two food preservatives: Sodium benzoate and potassium benzoate. Food Chem. Toxicol. 49(4):763- 769.

[9] Yadav, A., Kumar, A., Das, M. and Tripathi, A. 2016. Sodium benzoate, a food preservative, affects the functional and activation status of splenocytes at non cytotoxic dose. Food Chem. Toxicol. 88:40-47.

[10] Thomas, O.E. and Adegoke, O.A. 2015. Toxicity of food colours and additives: A review. Afr. J. Biotechnol. 9(36):900-914.

[11] Ramalho, V.C. and Jorge, N. 2006. Antioxidants used in oils, fats and fatty foods. Quím. Nova. 29(4):755-760.

[12] Noorafshan, A., Erfanizadeh, M. and Doust, S.K. 2014. Stereological studies of the effects of sodium benzoate or ascorbic acid on rats' cerebellum. Saudi Med. J. 35(12):1494-1500.

[13] Martyn, D.M., Mcnulty, B.A., Nugget, A.P. and Gibney, M.J. 2013. Food additives and preschool children. Proc. Nutr. Soc. 72(1):109-116.

[14] Michaelsson, G. and Juhlin, L. 1973. Urticarial induced by preservatives and dye additives in foods and drugs. Br. J. Dematol. 88(6):525-532.

[15] Nair, B. 2001. Final report on the safety assessment of benzyl alcohol, benzoic acid, and sodium benzoate. Int. J. Toxicol. 20(3):23-50.

[16] Boris, M. and Mandel, F.S. 1994. Foods and additives are common causes of attention deficit hyperactivity disorder in children. Ann. Allergy.72(5):462-468.

[17] Bridges, J.W., French, M.R., Smith, R.L. and Williams, R.T. 1970. The fate of benzoic acid in various species. Biochem. J. 118(1):47-51.

[18] Ren, L., Meng, M., Wang, P., Xu, Z., Eremin, S.A., Zhao, J., Yin, Y. and Xi, R. 2014.Determination of sodium benzoate in food products by fluorescence polarization immunoassay. Talanta 121:136-143.