

DETERMINATION OF CONCENTRATION OF SODIUM BENZOATE IN JUICES**Ritu Gupta* and Rakesh Kumar Yadav****Department of Chemistry, Dr. C.V. Raman University, Kargi Road Kota, Bilaspur (C.G.)***Corresponding Author's E mail: ritugu795@gmail.com

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ABSTRACT

Food preservatives are the substance added to food products to prevent decomposition by microbial growth. Benzoic acid is a commonly used antimicrobial preservative in food and beverages, especially in carbonated beverages. Excessive intake of benzoic acid may cause diarrhoea, abdominal pain and other symptoms. Thus, the maximum allowed concentration of Benzoic acid every kind of food are restricted by legislation. According to the Food and Drug Administration, the restriction of Benzoic acid is kept as 1000 mg/kg. An experimental study was performed by High Performance Liquid Chromatography for determining the concentration of sodium benzoate in juices available in market.

Keywords: Food preservatives, antimicrobial, carbonated beverages, diarrhoea, and legislation.

INTRODUCTION**Background**

Chemical food preservatives are widely used in the food industry and are invariably cheap ingredients that are effective against a wide range of spoilage organisms. Chemical food preservatives include compounds such as sodium benzoate, benzoic acid, nitrites, sulphites, sodium sorbate and potassium sorbate. Food preservatives are the substances added to foods to inhibit the growth of microorganisms. Majority of food available in the market contains different types of additives, among which preservatives are playing an important role.¹

Benzoic acid (BA) is a commonly used antimicrobial preservative in food and beverages, especially in carbonated beverages, as it presents its strongest antibacterial activity at pH 2.5–4.0. BA has inhibitory effects on the proliferation of bacteria and yeasts, a major cause of food spoilage. Benzoic acid is an organic compound which is described by chemical formula C_6H_5COOH . It consists of a carboxylic group attached to benzene ring. This compound exists as a crystalline colourless solid under normal condition.²

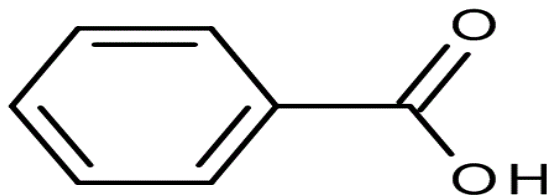


Fig 1: Benzoic acid

Benzoic acid and its sodium salt are used as preservatives, which are mainly used in pickled products and beverages. The maximum amount used in foods ranges from 0.2 to 2.0 g/kg. If benzoic acid is excessively added to the food, it will destroy the VB1 in the food and make the calcium insoluble, which can destroy the absorption of calcium by the human body. Furthermore, a long-term intake of benzoic acid will increase the risk of cancer. Therefore, it is necessary to ensure low levels of these preservatives in food to meet regulatory standards.³

Side effects of benzoic acid

Benzoic acid is generally recognized as safe by the Food and Drug Administration (FDA) when used in food amounts, with a maximum usage set at 0.1 percent for food products. However, there are still several benzoic acid hazards to consider, especially when this compound becomes a regular part of our diet. For instance, several studies have found that sodium benzoate, which is the sodium salt of this acid, may be tied to increased hyperactivity.⁴

Many people also commonly wonder: Is benzoic acid a carcinogen? Although the acid itself is not carcinogenic, it can form benzene when combined with vitamin C. Benzene is a compound that has been associated with a higher risk of cancer development. Carbonated beverages, in particular, contain the highest amount of benzene. Some research suggests that sugar-free beverages may be more prone to benzene formation. Therefore, it's best to limit consumption of processed foods that contain this compound in any of its forms. Not only are they likely pumped full of other fillers and additives that could potentially be harmful to health, but they're also typically high in sodium, added sugar and other unhealthy ingredients as well.⁵

Additionally, this acid has been linked to several adverse side effects in some people. It may cause irritation and inflammation. It may also trigger food allergy symptoms in certain people, including

rashes, itching and swelling. If you experience these or any other adverse side effects, it's recommended to discontinue use and consult with your doctor.⁶

1. MATERIALS AND METHODS

Material

All chemicals and reagents used in this study were of analytical or HPLC grade. HPLC-grade acetonitrile, used in the preparation of the mobile phase, was procured from Merck India Pvt. Ltd., Mumbai, Maharashtra. Potassium dihydrogen orthophosphate (KH_2PO_4) and orthophosphoric acid used for buffer preparation were purchased from SD Fine-Chem Limited, Mumbai, Maharashtra. Sodium benzoate standard (purity $\geq 99\%$) was procured from Loba Chemie Pvt. Ltd., Mumbai, Maharashtra.

Various commercial fruit juices and soft drink samples were collected from local retail outlets in Bilaspur, Chhattisgarh, and stored at 4°C until analysis. All filtration processes used $0.20\ \mu\text{m}$ nylon syringe filters obtained from Axiva Sicheem Biotech, New Delhi, India. The centrifuge used for sample clarification was manufactured by Remi Instruments, Mumbai, Maharashtra. The HPLC system used for analysis was an Agilent 1100 series system equipped with a UV detector.

Procedure

Basic principle of High-Performance Liquid Chromatography (HPLC): A kind of column chromatography known as High Performance Liquid Chromatography (HPLC) involves pumping a sample mixture or analyte in a solvent (mobile phase) through a column that includes chromatographic packing material (stationary phase) at a high pressure.⁷ The HPLC separation process is dependent on the distribution. Sample distribution between a stationary phase (column packing material) and a mobile phase. The analyte chemical structure determines whether the molecules are retarded during the stationary-phase transition. Detectors classify the UV, VIS, and PDA detectors according on absorption. They offer light-absorbing compounds high sensitivity. A widely popular detector for HPLC analysis is the UV detector.⁸

Sample preparation:

Three steps were involved in the HPLC analysis of sodium benzoate in various food samples.

Reagent Preparation:

To prepare the mobile phase, 250 mL of HPLC-grade acetonitrile was measured and transferred into a 500 mL volumetric flask. The volume was then made up to the mark with deionized water, and the

flask was shaken thoroughly to ensure complete mixing. This yielded a 50% (v/v) acetonitrile solution, which was used as the mobile phase for HPLC analysis.⁹

Sodium Benzoate Extraction from Juices and Soft Drinks:

Approximately 25 grams of each fruit product or soft drink sample was accurately weighed, homogenized using a blender, and transferred into a clean beaker. To this, 50 mL of distilled water was added, and the mixture was stirred thoroughly to ensure uniform extraction. The mixture was then filtered, and 20 mL of the resulting filtrate was subjected to centrifugation at 10,000 rpm for 10 minutes. From the clear supernatant, 10 mL was further diluted with potassium dihydrogen orthophosphate buffer to a final volume of 50 mL. A 30 μ L aliquot of this diluted solution was prepared for HPLC injection.¹⁰

Filtration:

The prepared sample solution was filtered through a 0.20 μ m syringe filter to remove any remaining particulates. The initial few drops of the filtrate were discarded to ensure cleanliness, and 1 mL of the subsequent filtrate was collected into an injection vial. This final filtered solution was then used for analysis via the HPLC system.¹¹

Mobile phase preparation:

Preparation of the mobile phase 0.05M In order to prepare the potassium dihydrogen orthophosphate buffer, 6.8045g of potassium dihydrogen orthophosphate was dissolved in 1000ml of distilled water, and its pH was adjusted to 2.5 using concentrated orthophosphoric acid. 700ml of the buffer was filtered and degassed, and 300ml of ACN was combined with it to be used as the mobile phase for HPLC analysis.¹²

Preparation of standard solutions:

HPLC standards 0.100g of sodium benzoate standards was weighed accurately and transferred to 100ml volumetric flask, dissolved in water by shaking and made up to the volume to yield 1000ppm sodium benzoate solution. Serial dilution using the equation $C_1V_1=C_2V_2$ was done using phosphate buffer to top up to the mark to 50ml to yield 10ppm, 20ppm, 40ppm, 60ppm, 80ppm and 100ppm respectively. These working standards were micro filtered and injected into HPLC and run at 230nm.¹³

Calculation

$$(A_2 \times C_1 \times DF) / A_1$$

C_1 = Concentration of standard

A_1 = Peak area of standard

A_2 = Peak area of sample

DF = Dilution factor

RESULT AND DISCUSSION

Chromatograms are graphical representations of the signal versus time. They display peaks, which are components of the sample that emerge at their corresponding times, which are corresponding times, which are referred to as retention times.

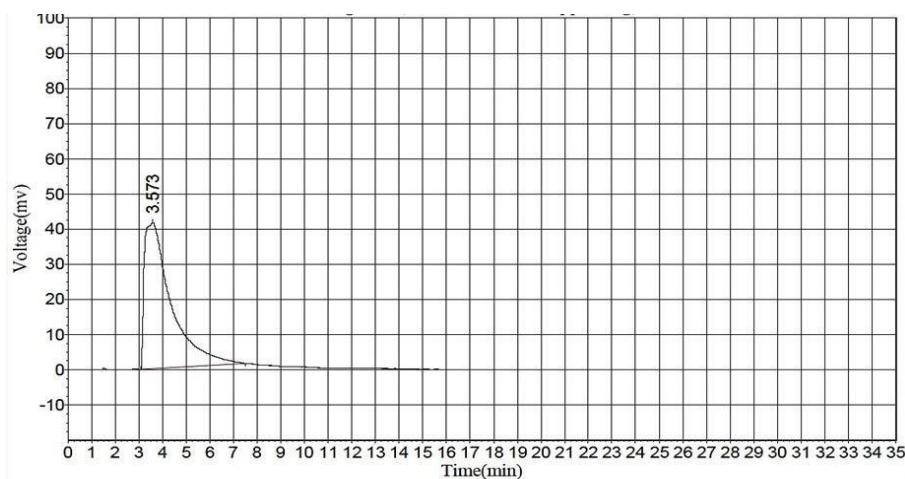


Fig 2: Chromatogram of standard Sodium Benzoate

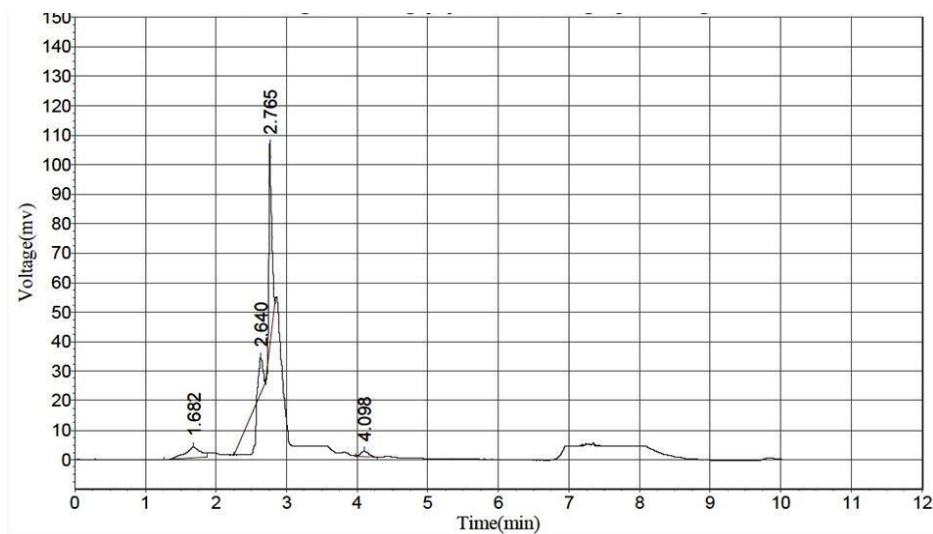


Fig 3: Chromatogram of orange juice

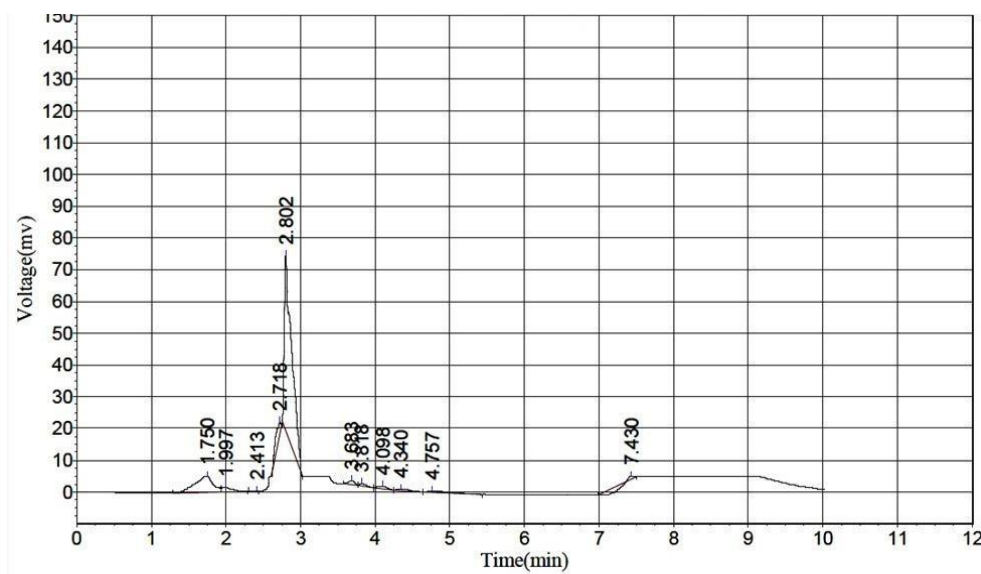


Fig 4: Chromatogram of mango juice

Table 1: Concentration of Sodium benzoate in Juices

Sample name	Concentration of sodium benzoate
Orange juice	699.552ppm
Mango juice	787.631ppm

It was discovered that the concentration of sodium benzoate was 699.552 parts per million and discovered that the concentration of sodium benzoate was 787.631 parts per million. The concentration of mango juice (787.631ppm) was found to be higher than that of orange juice, which was found to be 699.552 parts per million. It was discovered that. Each level of the preservative (sodium benzoate) tested were compliance with the food standard regulation of WHO and FDA (1000ppm).¹⁴

CONCLUSION

Food preservatives are substances that are added to food to prolong its shelf life and prevent spoilage. Ingesting high amounts of some preservatives or using them over an extended period of time may have negative health impacts, even though they play a vital function in preventing food borne illnesses and reducing food waste. The following are a few detrimental effects of food preservatives:

Preservatives are used to increase the shelf life of food and to maintain the quality for longer time chemical food preservatives reacts with cellular component of the body leading to the various food disturbance. If we must use food preservatives because of its advantage they should be generally recognised its safe extractable daily intake should not be exceeded. To minimise risk of health problems before purchasing canned food its ingredient should be checked

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