Analysis of Sodium Benzoate Preservative in Soft Drinks using Prominence-i HPLC

A rapid conventional HPLC method is described for the determination of food preservative - sodium benzoate in soft drinks. The method utilizes the new integrated Shimadzu HPLC Prominence-i LC-2030C.

In today’s world, the role of food additives becomes inevitable due to the increase in the production of treated and ready-to-eat convenience foods. Food additives are intentionally added to the food stuffs and must be safe for lifetime consumption. They are used for the purpose of maintaining or improving the quality, texture, consistency, appearance of the food products. Food additives are classified on the basis of their functional use, in which the preservatives are most important additive and are used to prevent and retard the microbial spoilage of food. Preservatives are classified into two types; - Class I and Class II.

The Class I preservatives are common salt, sugar, glucose, dextrose, spices, vinegar, honey, etc. The Class II preservatives are nature identical / synthetic compounds like salts of benzoic acid, sorbic acids, lactic acid, etc. The addition of Class I preservatives are not restricted in food products, but the class II preservatives are Generally Recognized As Safe (GRAS) and are listed in FDA GRAS food substances. It is noted that, the benzoic acid and sodium benzoate may be used as antimicrobial and flavouring agents in food products. The limit of benzoic acid / sodium benzoate as per FDA is 0.1% in food products.

Though the legislations are permitting these class II preservatives in food products, their use demands special care. In this context, a rapid and simple HPLC method has been developed and demonstrated in this article by IICMS using the new integrated Shimadzu HPLC ‘Prominence-i’ LC-2030C to quantitatively determine the content of sodium benzoate in grape, lemon and orange flavoured soft drinks.

Experimental

Preparation of Sodium benzoate standard

Prepared the concentration of 100mg/L sodium benzoate in diluent and used as working standard.

Preparation of Sample solution

The grape, lemon & orange flavoured soft drink samples with Class-II preservative were selected in this study. These soft drink samples were degassed thoroughly in ultrasonic bath for 20min. The sample aliquot of 0.5g of soft drink was transferred to 5mL volumetric flask & made up to the mark with diluent and sonicated to mix well.

Results and Discussion

The sodium benzoate standard solution and the simply diluted soft drink samples were injected directly into the Prominence-i LC-2030C chromatography system without any complex sample preparation and recorded the results. The retention time of sodium benzoate is observed about in 3.5min. The typical

Table 1: Instrument and Analytical Conditions

<table>
<thead>
<tr>
<th>HPLC</th>
<th>Prominence-i LC-2030C</th>
</tr>
</thead>
<tbody>
<tr>
<td>Column</td>
<td>C-6 Phenyl Hexyl (50x4.6mm, 3µ)</td>
</tr>
<tr>
<td>Workstation</td>
<td>Lab solution Version 5.62 P</td>
</tr>
<tr>
<td>Flow Rate</td>
<td>0.7mL/min (Isocratic)</td>
</tr>
<tr>
<td>Mobile Phase</td>
<td>10mM Ammonium formate, pH adjusted to 3.0 with dilute formic acid : Acetonitrile (75:25)</td>
</tr>
<tr>
<td>Diluent</td>
<td>Water : Acetonitrile (75:25)</td>
</tr>
<tr>
<td>Wavelength</td>
<td>230nm</td>
</tr>
<tr>
<td>Injection Volume</td>
<td>5 µL</td>
</tr>
<tr>
<td>Run Time</td>
<td>7.00 minutes</td>
</tr>
</tbody>
</table>

Fig 1: Structure of Sodium Benzoate

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HPLC chromatogram of sodium benzoate standard 100mg/L concentration is shown below:

The method is tested for the HPLC validation parameters and found to be precise, linear and accurate. The %RSD of area response for the six replicate sodium benzoate standard injections is found to be 0.25%. The area response is tabulated in Table 2: Area response of Sodium Benzoate 100mg/L standard.

### Table 2: Area response of Sodium Benzoate 100mg/L standard

<table>
<thead>
<tr>
<th>Injection</th>
<th>RT (min)</th>
<th>Area Response</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>3.55</td>
<td>2970435</td>
</tr>
<tr>
<td>2</td>
<td>3.55</td>
<td>2963829</td>
</tr>
<tr>
<td>3</td>
<td>3.55</td>
<td>2964031</td>
</tr>
<tr>
<td>4</td>
<td>3.55</td>
<td>2961078</td>
</tr>
<tr>
<td>5</td>
<td>3.55</td>
<td>2955879</td>
</tr>
<tr>
<td>6</td>
<td>3.56</td>
<td>2948774</td>
</tr>
<tr>
<td>Average</td>
<td>3.55</td>
<td>2960671</td>
</tr>
<tr>
<td>STDEV</td>
<td>0.0041</td>
<td>7500.2104</td>
</tr>
<tr>
<td>%RSD</td>
<td>0.11</td>
<td>0.25</td>
</tr>
</tbody>
</table>

The grape flavoured soft drink was taken for this validation study. The %RSD for the content of sodium benzoate for the six replicate preparation of grape flavoured soft drink was found to be 1.05%. The data and overlaid HPLC chromatograms are shown in Table 3 and Fig.3 respectively.

The method is found to be linear between 2.0mg/L to 200mg/L of sodium benzoate standard. The linearity graph was plotted between the concentration and the area response of sodium benzoate. The linearity graph was plotted between the concentration and the area response of sodium benzoate.