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COMMUNICATION I

Determination of Sugars in Soft Drinks by High Performance Liquid Chromatography

Key words: HPLC, soluble sugars; soft drinks.

ABSTRAK

Kandungan fruktosa, glukosa dan sukrosa dalam minimum ringan terpilih yang terdapat di Malaysia ditentukan melalui kromatografi cecair prestasi tinggi (HPLC). Minuman ringan yang diuji mengandungi 8.5 – 15.3 g 100 ml $^{-1}$ gula larut. Kandungan fruktosa, glukosa dan sukrosa masing-masing didapati dalam julat 0 – 6.7, 0 – 6.9 dan 0 – 10.5 g 100 ml $^{-1}$. Didapati bahawa pada minumam ringan yang spesifik, perbezaan adalah lebih besar antara kandungannya untuk gula individu daripada kandungannya bagi jumlah gula.

ABSTRACT

The fructose, glucose and sucrose contents of selected soft drinks available in Malaysian markets were determined by high performance liquid chromatography (HPLC). The soft drinks tested had a soluble sugar content of between 8.5 to 15.3 g 100 ml $^{-1}$. The average fructose, glucose and sucrose contents were found to be in the ranges of 0 – 6.7, 0 – 6.9 and 0 – 10.5 g 100 ml $^{-1}$ respectively. The content of individual sugars were found to be more variable than the content of total sugar in different samples of a specific soft drink.

INTRODUCTION

The past few years has seen the mushrooming of the soft drink industry in Malaysia. Besides the well known soft drinks like Seven Up and Coca-cola, many new varieties which are lesser known in the West, have gained popularity in the local market. These include chrysanthenum and herbal tea, longan-winter melon and sugar cane drinks, as well as fruit juices like guava, mango and mango-pineapple juice.

Interest in the sweetened soft drinks, both carbonated and non-carbonated, arises from two considerations. Firstly, the analysis of the sugar contents of locally produced soft drinks by high pressure liquid chromatography (HPLC) has not been undertaken before. Secondly, from a nutritional view point, sucrose intake at high levels has been implicated in such health problems as diabetes mellitus (Cohen *et al.*, 1974), atherosclerotic heart disease (Kaufmann *et al.*, 1967), carbohydrate malabsorption syndromes (Donaldson and Grybosky, 1973) and dental caries (Hartles, 1967).

MATERIALS AND METHODS

Materials

Samples of soft drinks were bought from local supermarkets. 4 samples of each drink were analysed. The drinks selected were: (i) Orange crush (F & N), (ii) Coca-cola, (iii) A & W root beer, (iv) Dads root beer, (v) Schweppes orange, (vi) Fanta (Gedep Merk) vruchtenlimonade sinaasappel, (vii) Yeo's longan-winter melon drink, (viii) Drinho chrysanthenum tea (ix) Drinho sugar cane drink, (x) Joy mango juice (xi) Joy guava juice, (xii) Delite mango juice (xiii) Dewi mango juice, (xiv) Sunjus orange juice (xv) Green Spot orange juice drink and (xvi) Drinho herbal tea.

Analyses of Sugars

A HP 1048B liquid chromatograph with an RI detector was used. The method was based on that of Hurst *et al.*, 1977. The column was an NH_2 polar bonded phase column, 10 μm (250 mm × 4.6 mm I.D.). The mobile phase was

acetonitrile:water (85:15) and the flow rate was 2.5 cm min $^{-1}$. The injection volume was 10 μl. Identification and quantification of sugars were done by comparing retention times and peak areas of samples to peak areas of standards as peak area was directly proportional to the concentration of the standard throughout the concentration range used.

Soft drinks were analysed as received with minor sample preparation. Carbonated drinks were decarbonated by vigorous agitation of solutions with a glass rod for 5 min. For the analysis, 2 ml of the drink was diluted to 10 ml in a volumetric flask. The diluted drink solution was filtered through C-18 Sep Pak cartridges and 0.45 μm filters prior to injection into the HPLC. Recovery studies were carried out by spiking soft drinks with known amounts of a standard sugar solution containing glucose, fructose and sucrose. For spiking experiments, 2 ml of standard sugar solution containing 1.6×10^{-2} g ml $^{-1}$ of each of the sugars glucose, fructose and sucrose was mixed with 1 ml of the undiluted drink and the mixture was made up to volume in a 10 ml volumetric flask. These studies showed that the recovery for fructose, glucose and sucrose for the range of soft drinks was between 91 to 107%. However, the variation in the % recovery between individual sugars in a specific drink was much less and averaged about 2.6%.

RESULTS AND DISCUSSION

Sugar Content of Soft Drinks

Fig. 1 shows two chromatograms obtained using the HPLC conditions described. Table 1 shows the data obtained for the soft drinks analysed. The total sugar content was found to be between 8.5 to 15.3 g 100 ml $^{-1}$. The lowest sugar content was found in Drinho sugar cane drink while the highest was found in one batch of Joy guava juice. The average value for total sugar in the drinks analysed in this work was found to be 12 g 100 ml $^{-1}$. Vidal-Valverde *et al.* (1985) found that the total amount of sugars ranged from 9.2 to 14 g 100 ml $^{-1}$ in soft drinks and fruit nectars he analysed. A total sugar content of 10 to 12 g 100 ml $^{-1}$ was found in most

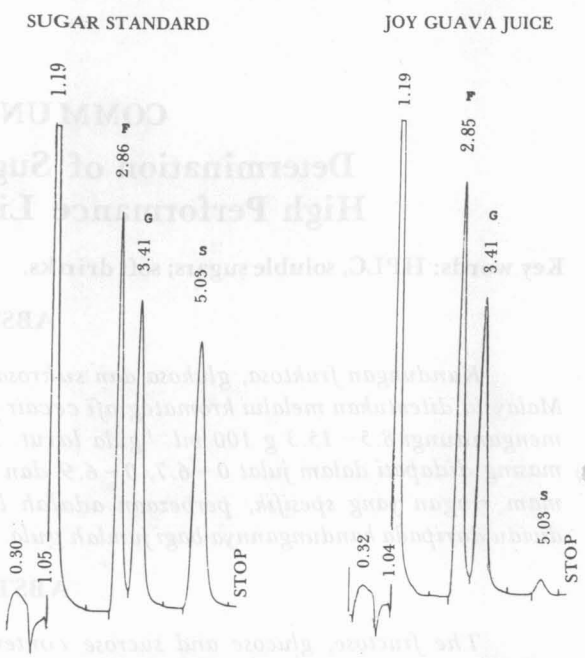

Fig. 1: HPLC chromatograms of (i) standard sugar solution and (ii) diluted Joy guava juice. (F — fructose; G — Glucose; S — sucrose)

drinks analysed by Martin-Villa *et al.* (1981). Thus, it is evident that the range of total sugars in soft drinks found in locally available soft drinks is in a similar range to that reported. There are however more marked differences in the contents of individual sugars.

There is a wide variation in the content of individual sugars present in the range of soft drinks analysed. The average fructose and glucose contents were found to be 0 – 6.7 and 0 – 6.9 g 100 ml $^{-1}$ respectively. The average sucrose content was in the range of 0 – 10.5 g 100 ml $^{-1}$. One of the main reasons for the differences in the content of individual sugars is the different amounts of corn sweeteners and sucrose used in the manufacture of different drinks. Labels on the soft drink containers indicated that in some drinks corn sweeteners (which includes high fructose-glucose syrups) or/and sugar was used in the manufacture of the drinks. The labels on some of the other drinks stated only that 'sugar' was used as an ingredient. The definitions of sugar and sweeteners for soft

TABLE 1
Soluble sugars in soft drinks

Type of Drink	Container	Fructose g 100 ml $^{-1}$	Glucose g 100 ml $^{-1}$	Sucrose g 100 ml $^{-1}$	Total Sugars g 100 ml $^{-1}$
Orange Crush (F&N)	Bottle	6.57 ± 0.09	6.31 ± 0.23	< 0.46	13.00 ± 0.41
Coca-Cola	Bottle	5.65 ± 0.57	5.57 ± 0.05	< 0.16	11.35 ± 0.12
	Can	3.40 ± 0.25	3.65 ± 0.25	3.60 ± 0.09	10.65 ± 0.43
A & W root beer	Can	6.62 ± 0.12	4.71 ± 0.11	n.d.	11.32 ± 0.23
Dads root beer	Can	3.75 ± 0.10	3.74 ± 0.09	4.25 ± 0.52	11.74 ± 0.35
Schweppes orange	Can	6.58 ± 0.28	6.36 ± 0.24	< 1.19	13.41 ± 0.38
Fanta (Gedep Merk) vruchtenlimonade sinaasappel	Can	6.69 ± 0.44	6.50 ± 0.41	< 0.91	13.67 ± 0.92
Yeo's longan-winter melon drink	Tetra-pak	0.43 ± 0.02	0.42 ± 0.02	9.59 ± 0.46	10.45 ± 0.47
Drinho chrysanthenum tea	Tetra-pak	< 0.06	< 0.05	10.48 ± 0.50	10.53 ± 0.50
Drinho sugar cane drink	Tetra-pak (a)	1.41 ± 0.01	1.97 ± 0.01	5.10 ± 0.01	8.47 ± 0.01
	(b)	0.58 ± 0.09	0.55 ± 0.12	9.19 ± 0.18	10.20 ± 0.35
Joy mango juice	Tetra-pak	3.53 ± 0.13	3.11 ± 0.08	5.64 ± 0.14	12.28 ± 0.32
Dewi mango juice	Tetra-pak (a)	6.23 ± 0.15	6.28 ± 0.44	2.42 ± 0.34	14.93 ± 0.93
	(b)	5.60 ± 0.17	5.01 ± 0.11	2.10 ± 0.87	12.72 ± 0.32
Delite mango-pineapple drink	Tetra-pak	6.04 ± 0.16	6.06 ± 0.23	< 1.47	12.64 ± 0.16
Sunjus orange juice	Tetra-pak	6.62 ± 0.11	6.48 ± 0.08	< 1.22	14.05 ± 0.16
Joy guava juice	Tetra-pak (a)	4.46 ± 0.35	5.27 ± 0.26	5.58 ± 0.56	15.31 ± 0.05
	(b)	6.39 ± 0.10	6.16 ± 0.66	0.31 ± 0.02	12.82 ± 0.11
Green Spot orange juice drink	Tetra-pak (a)	6.41 ± 0.18	6.93 ± 0.46	0.82 ± 0.25	14.44 ± 0.98
	(b)	6.22 ± 0.09	6.08 ± 0.20	< 0.57	12.67 ± 0.07
Drinho herbal tea	Tetra-pak	< 0.03	< 0.03	9.81 ± 0.20	9.83 ± 0.18

The symbols (a) and (b) refer to analyses of different batches of drinks which were bought at different times. In drinks, other than those with symbols (a) and (b), the differences in sugar contents between batches were small and less marked. n.d. = not detectable.

drinks vary from country to country. In the 1964 Soft Drink Regulation (UK), sugar is defined as 'any soluble carbohydrate sweetening matter' while the sugar product regulations of the EEC identifies the term sugar with 'sucrose' (Tilley, 1978). In the Food Regulations 1985 of Malaysia sugar is defined as the food chemically known as sucrose and includes granulated sugar, loaf sugar, castor sugar and powdered sugar. Sugar should contain not less than 99.5% of sucrose.

The differences in the content of individual sugar can also arise because of other factors. In fruit juice drinks where the juice, concentrate or puree of the fruit is added as an ingredient, differences in the contents of individual sugars in the drink may be due to inherent differences in the contents of individual sugars in different fruits. Another contributory factor is the inversion of sucrose during storage. It has been stated that it is quite reasonable to assume that sucrose undergoes hydrolysis in the acidic pH of soft drink media (Martin-Villa *et al.*, 1981; Vidal Valverde *et al.*, 1985).

In this work, there were also marked differences in the individual sugar content in different samples of Coca-cola, Drinho sugar cane drink, Dewi mango juice, Joy guava juice and Green Spot orange juice drink. The pH of these drinks was found to be between 2.5 to 4.8. It would appear that these differences may be partly due to inversion of sucrose during storage. In the paper by Vidal-Valverde *et al.*, (1985) it was also stated that a low sucrose content was probably an indication of long storage time. A number of reports have shown that there can be significant differences in the individual sugar content of the same type of drink. Pinalla (1968) found 9.5% sucrose in cola drinks while Southgate *et al.*, (1978) found 0.5% sucrose in cola drinks. In recent analysis, the sucrose level in Coca-cola was found to vary from 3.3 to 7.3 g 100 ml $^{-1}$ in 3 different samples of Coca-cola drinks while the individual sugar contents of some canned lemon, orange, pineapple drinks and Seven-up, Sprite and Tonic water were also found to be variable (Vidal-Valverde *et al.*, 1985). In addition to differences in contents of individual sugars, there were also differences in the content of total soluble sugars. This was found most prominently in Drinho sugar cane drink, Dewi mango juice, Joy guava juice and Green Spot orange drink. These differences are probably due primarily to different production batches of the drinks. Variations in sugar content of this range has also been reported by other workers (Vidal-Valverde *et al.*, 1985).

Nutritional Implications

The analysis revealed that the usual packet of soft drink (250 ml) contains between 21 to 38 g of sugar. This constitutes a significant level considering that the average daily intake of sugar by Malaysians is nearly 100 g (FAO, 1984). Since soft drinks are popular among children, concern is expressed for the role soft drinks may have in contributing to dental caries. Among the health problems related to high intakes of refined carbohydrates, dental caries is most clearly linked to the quantity and frequency of sufar ingested (Bierman, 1979).

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