

Evaluation of Sugar, HMF, pH, and Acidity Content in Some Popular Soft Drinks Sold in Türkiye

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Abstract: It is well-known that high consumption of sugary drinks causes negative effects on human health. This study aims to determine the sugar, HMF, pH, and titratable acidity values of soft drinks (n = 25) sold in Türkiye. All the samples were purchased from chain supermarkets in the city of Bingöl, Türkiye. HPLC-RID and HPLC-DAD were used to determine sugar profile (glucose, fructose, sucrose, and maltose) and HMF content of the samples, respectively. Results showed that the total sugar and HMF contents of the samples were found to range from 0.23 to 13.85 % and from 0.65 to 27.76 mg/L respectively. In 9 out of 25 samples HMF were not detected. Furthermore, the pH and acidity values of the samples were found to vary from 1.96 to 6.34 and from 0.73 to 22.07 mEq/L, respectively. The samples were classified based on their total sugar content (from sugar free to very high sugar) and pH erosive capacity (from minimally erosive to extremely erosive). Most of the samples (15 out of 25 samples) were classified in the high sugar content soft drinks (>8g/100ml) and 14 out of 25 samples were found to be considered as extremely erosive (pH<3).

Türkiye'de Satılan Bazı Popüler Meşrubatların Şeker, HMF, pH ve Asitlik İçeriğinin Değerlendirilmesi

Anahtar Kelimeler

Eroziv etki, HMF, Halk sağlığı, Meşrubatlar, Şekerler, Şeker vergisi

Öz: Şekerli içeceklerin yüksek tüketiminin insan sağlığı üzerinde olumsuz etkilere neden olduğu bilinmektedir. Bu çalışma, Türkiye'de satılan meşrubatların (n=25) şeker, HMF, pH ve titre edilebilir asitlik değerlerini belirlemeyi amaçlamaktadır. Tüm örnekler, Türkiye'nin Bingöl ilindeki zincir süpermarketlerden satın alınmıştır. Numunelerin şeker profilini (glukoz, fruktoz, sukroz ve maltoz) ve HMF içeriğini belirlemek için sırasıyla HPLC-RID ve HPLC-DAD kullanılmıştır. Elde edilen sonuçlara göre, örneklerin toplam şeker ve HMF içeriklerinin sırasıyla % 0,23 - % 13,85 ve 0,65 - 27,76 mg/L arasında değiştiği belirlenmiştir. 25 örneğin 9'unda HMF tespit edilmiştir. Ayrıca örneklerin pH ve asitlik değerleri sırasıyla 1,96 - 6,34 ve 0,73 - 22,07 mEq/L arasında olduğu tespit edilmiştir. Numuneler, toplam şeker içeriklerine (şekersizden - çok yüksek şeker) ve pH eroziv kapasitelerine (minimum erozivden - aşırı erozive) göre sınıflandırılmıştır. Örneklerin çoğu (25 örnekten 15'i) yüksek şeker içerikli meşrubatlar (>8g/100ml) sınıfına girmiş ve 25 örnekten 14'ünün aşırı eroziv (pH<3) olarak değerlendirilebileceği saptanmıştır.

1. INTRODUCTION

Soft drinks comprise sugar sweetened beverages which are so called 'sugary drinks' and 'no added sugar' drinks that just contain naturally occurring sugars. The WHO (World Health Organization) defines sugary drinks as "all types of beverages containing free sugars and these include carbonated or non-carbonated soft drinks,

fruit/vegetable juices and drinks, liquid and powder concentrates, flavoured water, energy and sports drinks, ready-to-drink tea, ready-to-drink coffee, and flavoured milk drinks." The WHO in its report recommended that countries should implement fiscal policies to reduce the consumption of sugary drinks [1]. Hence, some countries have made legal arrangements taking into account the WHO's suggestion. For example, the government of the UK brought the Soft Drinks Industry Levy into force in

2018. According to this levy, companies will be extra charged 24p/L, and 18p/L respectively if soft drinks include added sugars ≥ 8 g / 100 mL, and between ≤ 8 g / 100 mL - ≥ 5 g / 100 mL. [2]. Similar taxations in different rates on sugar added soft drinks have been implemented in France, the Philippines, Chile, Mexico, Switzerland [3]. Furthermore, regardless of amount of sugar content of sugar added soft drinks are subject to excise duty between 10 % - 35 % rates in Türkiye. According to the law, coke drinks are subjected to 35 % and other soft drinks including fruit juices, vegetable juices, mineral waters, sodas, and non-alcoholic drinks are subjected to 10% special consumption tax. These extra costs are paid directly by the consumers [4].

Soft drinks are very popular especially among children due to the taste, relatively low cost and preferred to be consumed at any time in any place; with meal, between meals. They are also a source of hydration and allow for quenching consumer's thirst [5, 6]. Turkish Statistical Institute [7] announced that children constitute 26.9 % of Türkiye's population (84 million 680 thousand 283). According to data obtained from Statista, 2377 million litres of soft drinks equivalent to 27.95 litres per person were consumed in Türkiye in 2021 [8]. In order to maintain a balanced and healthy diet, the WHO recommended that free sugar intake for adults and children should be less than 10% of daily energy intake [1].

There is a relationship among acidity, pH, sugar content, and HMF concentration. Soft drinks generally have low pH and high acidity values. The sources of acidity and low pH could be from their natural occurrence in

ingredients and from added acidulants. Low pH and high acidity eliminate microbial growth which occurs due to a high percentage of moisture and give beverages extra flavour [9, 10, 11]. Presence of HMF is an important quality indicator in many foods, although it is still not considered as a hazardous substance [12]. HMF has been identified in many food products including honey, syrups, and roasted foods such as coffee, malt, caramels [13]. Generally, its presence in soft drinks is an indicator for heat treatment. Heat treatment is an essential part of soft drink processing [14]. During thermal processing, in an aqueous solution with the presence of sugars, amino acids, and under acidic conditions high heating treatment for a certain time triggers to generate HMF [15].

The primary objectives of this study are: (i) to identify the sugar profile of the soft drinks and classify them depending on their total sugar content (from sugar free to very high sugar content) according to legislations; (ii) to analyse HMF content of the soft drinks; (iii) to determine pH and titratable acidity of the soft drinks and categorise them according to their erosive capacity.

2. MATERIAL AND METHOD

2.1. Material

Twenty-five soft drinks were purchased from a chain supermarket in city of Bingöl, Türkiye in January 2021. The selected chain supermarket has branches in all cities. Thus, beverages are available to be purchased by consumers in all cities of Türkiye. Type of samples and some of the labelling information were demonstrated in Table 1.

Table 1. Type of samples and some of the labelling information

Sample ID	Sample Type	Added sugar /syrup content	Acidulant content	Expiry Date/Batch No
SD1	Orange juice from concentrate	No sugar added	No acidulant added	19.07.2021/005016
SD2	Squeezed Orange juice	No sugar added	No acidulant added	11.08.2021/IZ-1112 0348813
SD3	Apricot juice from concentrate	Sucrose/fructose-glucose	Citric acid	26.11.2021/2611-IZ-M26
SD4	Grape juice from concentrate	Beet sugar	Citric acid	29.10.2021/2910-IZ-M25
SD5	Mango juice from concentrate	Beet sugar	Citric acid	24.04.2021/2404-IZ-M26
SD6	Pineapple juice from concentrate	Sucrose/fructose-glucose	Citric acid, trisodium citrate	04.11.2021/0411-IZ-M26
SD7	Apple juice from concentrate	Sucrose/fructose-glucose	Malic acid	15.03.2022/1512-IZ-M26
SD8	Squeezed apple juice	No sugar added	No acidulant added	17.07.2021/IZ-1711 000344
SD9	Squeezed pomegranate juice	No sugar added	No acidulant added	16.07.2021/IZ - 1611 020250
SD10	Cherry juice from concentrate	Beet sugar	Concentrated lemon juice	14.12.2021/1412 IZ-M22
SD11	Squeezed tomato juice	No sugar added	No acidulant added	07.10.2021/TT2047 071020
SD12	Lemonade soda	Sucrose	Concentrated lemon juice	25.08.2021/61211221
SD13	Lemonade soda	Sucrose/fructose-glucose	Citric acid, sodium citrate	11.05.2021/AA1612 111120-S
SD14	Mixed fruit flavoured soda	Sucrose/fructose-glucose	Citric acid	03.03.2021/0145CR03111203
SD15	Orange flavoured soda	Sucrose/fructose-glucose	Citric acid, malic acid, sodium citrate	29.04.2021/EN011 290720-S
SD16	Lemon flavoured Iced Tea	Sucrose	Citric acid, trisodium citrate	19.03.2022/14
SD17	Lemon flavoured Iced Tea	Sucrose/fructose-glucose	Citric acid, sodium citrate	17.04.2021/EN0204 170720-S
SD18	Lemon flavoured Iced Tea	Sucrose, fructose	Trisodium citrate, concentrated lemon juice	17.09.2021/0844AD 1709201(S)
SD19	Energy drink	Sucrose	Citric acid, sodium citrate	12.12.2022/EN0245 121220
SD20	Energy drink	Sucrose, glucose	Citric acid, sodium bicarbonate	27.11.2022/1879741
SD21	Energy drink	Sucrose/fructose-glucose	Citric acid, phosphoric acid, sodium bicarbonate	27.11.2021/AA0430 271120-S
SD22	Coke	Sucrose/fructose-glucose	Phosphoric acid	20.10.2021/DZ0037 201020-S
SD23	Coke	Sucrose/fructose-glucose	Ammonium sulfate	06.10.2021/1927CR0610201 (S)
SD24	Iced Coffee (Americano)	No sugar added	Sodium bicarbonate, tripotassium citrate	05/2021/02262226
SD25	Pineapple Flavoured Malt drink	Sucrose/fructose-glucose	Citric acid	06.11.2022/21.06 3H S

2.1. Method

2.2.1. Sugar profile analysis by HPLC- RID

2.2.1.1. Standard solution preparation

Sugar profile analyses of the samples were performed using the modified method of TS 13359 [16]. Stoke solution of fructose (D(-)-fructose, Sigma Aldrich), glucose (D-glucose anhydrous, Fluka), sucrose (Fluka), maltose (D-(+)- maltose monohydrate, Fluka) were prepared by dissolving them with ultrapure water (generated from Sartorius H2O-I-1-UV-T Arium Comfort; resistivity 18.2 MΩ.cm) in a beaker. The prepared solution was transferred to a 100 mL flask and then 25 mL methanol (Chromasolv grade, Sigma Aldrich) added to the flask. Later the flask was made up to mark by the addition of ultrapure water. By diluting the prepared stoke solution, seven points standard series was prepared to produce calibration curves. Calibration curves were created by plotting the area of the sugar compound against the concentration of the sugar compound. All the calibration curves showed good linear regressions in the range of 0.9993 – 0.9998 illustrated in Figure 1.

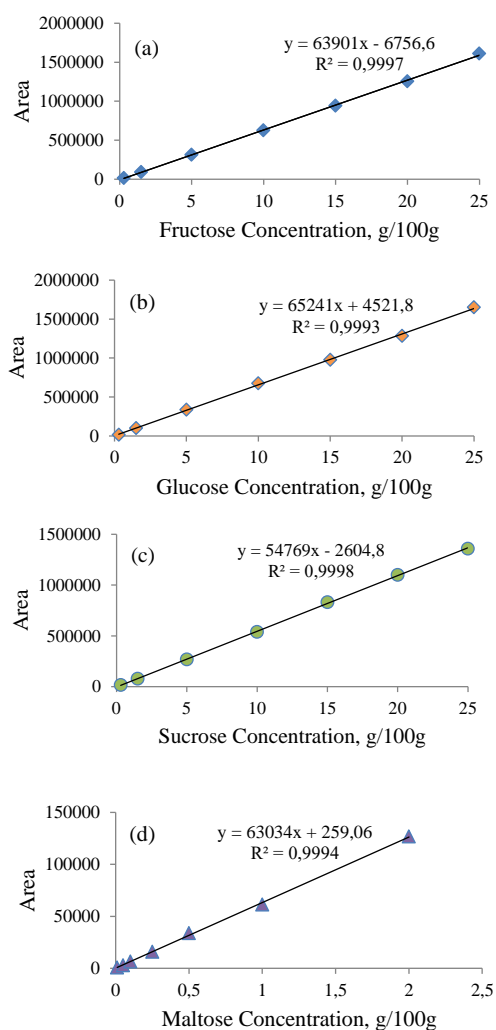


Figure 1. Calibration curves of: (a) fructose, (b) glucose, (c) sucrose and (d) maltose sugars standards

2.2.1.2. Sample preparation and data acquisition

5 g sample was weighed in a beaker and transferred to a volumetric 100 mL flask, and then 25 mL methanol was added to the flask. Later the flask was filled to the mark with ultrapure water and the flask was homogenised by shaking gently. The prepared solution was filtered through a 0.45 µm filter (Sartorius Stedim Biotech GmbH, Goettingen, Germany) into a 2 mL autosampler vial and then introduced to the HPLC instrument (Infinity II, Agilent Technologies) equipped with Refractive Index Detector (RID, 1260 Infinity II), using an EC 250/4.6 NUCLEODUR 100–5 NH2 RP column (250 mm, 4.6 mm, Macherey-Nagel) with EC HPLC guard column NUCLEODUR 100-5 NH2-RP (5 µm, 4x3 mm, Macherey-Nagel). Analysis was performed in isocratic elution mode with a mobile phase composed of acetonitrile (HPLC grade, Sigma Aldrich) - water (80:20 v/v) at a flow rate of 1.3 mL/min, injection volume of 10 µl at 30 °C column and detector temperature. Agilent LC Solutions software was used for data acquisition. All the measurements were done in triplicate.

2.2.2. HMF analysis by HPLC- DAD

2.2.2.1. Standard solution preparation

HMF content of the samples was performed by a modified IHC method [17]. In order to prepare 100 mg/L HMF Stock solution, 10 mg HMF standard (J&K, Haihang Industry Co., Ltd.) was weighed using micro analytical balance (XP6, Metler – Toledo). The weighted standard transferred in a 50 mL beaker and dissolved with 90:10 v/v water-methanol and then put into a 100 mL flask filled up with 90:10 v/v water-methanol. Later, the solution was diluted to obtain concentrations of 0.5 mg/L, 1 mg/L, 2 mg/L, 4 mg/L, 8 mg/L, 10 mg/L. 6 points standard series was prepared showing good linear regression ($R^2 = 0.9998$) given in Figure 2.

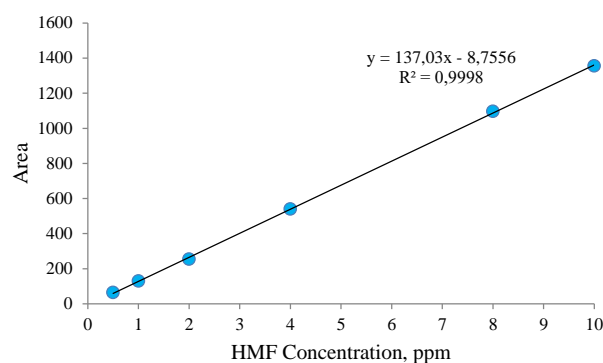


Figure 2. Calibration curve of HMF standard

2.2.2.2. Sample preparation and data acquisition

10 g of sample was weighed in a 50 mL beaker then dissolved with 40 mL ultrapure water. The solution was transferred in a 50 mL flask then 0.5 mL of Carrez I and 0.5 mL Carrez II solutions was added and was filled up 50 mL with ultrapure water. Prepared solution was shaken vigorously then held at ambient temperature for 14 hours. Supernatant was filtered through a paper filter

(MN 615 110 nm Macherey-Nagel). Later, the obtained solution was filtered through 0.45 µm filter (Sartorius Stedim Biotech GmbH, Goettingen, Germany) into a 2 mL autosampler then introduced to the HPLC instrument coupled with Diode Array Detector (DAD 1260 Infinity II WR) using an ACE 5 C18 (250 x 4.6mm id) column. The analysis was carried out in isocratic elution mode with a mobile phase composed of 90:10 v/v water-methanol. The flow rate was 1mL/min, a wavelength of 285nm, and 30 °C column temperature. Agilent LC solutions software was used for data acquisition. All the measurements were done in triplicate.

2.2.3. pH and titratable acidity

The pH was measured soon after opening the package. pH and titratable acidity were performed according to TS 1125 ISO 750. Firstly, pH meter (Orion 3-Star, Thermo Fisher Scientific Inc.) was calibrated using buffer solutions of pH 4, 7, and 10 (Thermo Scientific Inc.). Then, 25 mL of sample was added to a 250 mL beaker and placed on a magnetic stirrer. Later, the electrode was immersed into stirred sample solution and pH of the sample was noted at ambient temperature. The sample was then titrated with 0.1 mol/L NaOH (Sigma - Aldrich) to pH 8.1. Finally, titratable acidity was calculated as follows:

$$\text{Titratable acidity} = \frac{1000 \times V_1 \times C}{V_0} \quad (1)$$

where

V_1 : Volume of NaOH used for titration

C : Concentration of NaOH used for titration

V_0 : Total volume of the sample

All the measurements were done in triplicate.

3. RESULTS AND DISCUSSION

3.1. Sugar Analysis by HPLC- RID

Sugar profiles of the soft drinks including glucose, fructose, sucrose and maltose have been determined using HPLC-RID instrument. A typical sample chromatogram is given in Figure 3. The sugar concentration of the samples were calculated based on the peak areas of calibration curves of the standards by substituting the samples peak area.

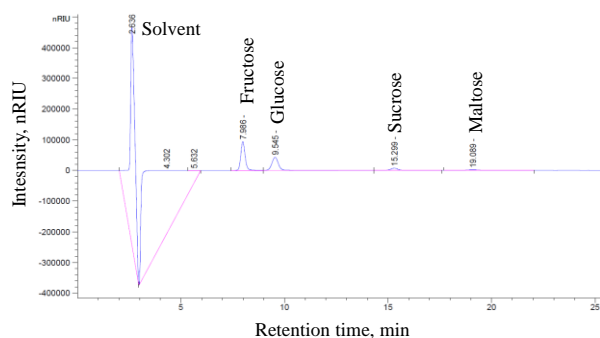


Figure 3. An example of a typical sample chromatogram for sugar components

Total sugar content of the samples were given in Table 2 and illustrated in Figure 4. Concentration of total sugar content of the samples ranged between 0.23 and 13.85 g/100 mL. The mean value of total sugar content of the samples was determined as 8.79 g/100 mL. Sucrose was found to be the most abundant sugar with the mean amount of 3.71 g/100 mL and maltose was found to be the least abundant sugar compound with the mean amount of 0.02 g/100 mL. Most frequently detected sugar components were fructose and glucose to be found in 24 out of 25 samples whilst maltose as least common sugar component were detected in only 3 samples. Unexpectedly, while 9.55% total sugar consisting of 1.02% fructose, 0.98% glucose, and 7.63% sucrose was detected in the sample of SD25 (pineapple flavoured malt drink), no maltose was found in the sample. The highest and the lowest total sugar concentration were found in SD19 (energy drink) sample and SD24 (iced coffee) sample as 13.22 and 0.23 g/100 mL, respectively. According to Nutrition and Health Claims Regulation (EC, 2006), soft drinks were classified in four levels according to their sugar content as sugar-free, moderate sugar content, high sugar content, and very high sugar content.

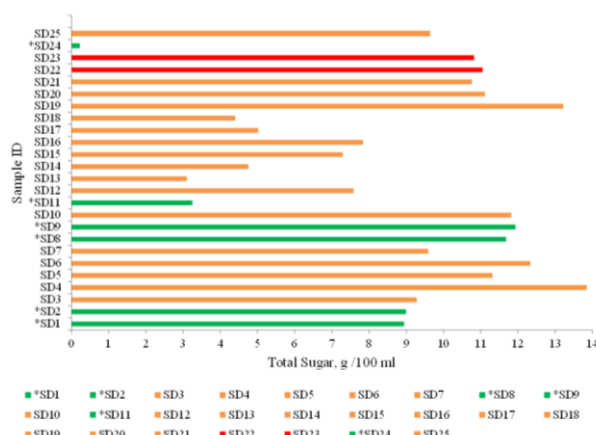


Figure 4. Total sugar content of the samples (green bars represent no taxation, orange bars represent 10% taxation, red bars represent 30% taxation, and '*' indicates the samples does not contain added sugar)

The evaluation of sugar content of the samples according to their sugar levels are illustrated in Figure 5. Most of the products (n=15) were to be determined in 'very high sugar' class. The rest number of 4, 5, and 1 sample were included in 'high', 'moderate' and 'free-sugar' classes, respectively.

Huizinga and Hubert [19] in their study analysed sugar content of the 463 soft drink products and resulted that 171 products were found to be in 'very high sugar' class, 103 products were included in the 'high sugar' class, 134 products were involved in 'moderate sugar' class, and 55 products were get involved in 'sugar-free' class. Silva et al. [20] stated that 48 out of 68 soft drinks including colas, juice drinks, iced teas, and flavoured drinks contained free sugars in their contents. The total sugar

contents including fructose, glucose and sucrose sugars were found ranging between 0.8 and 11 g/100 mL. They detected that colas and juice drinks contained higher total sugars comparing to other type of drinks. It was concluded that, after the sugar tax on soft drinks enforced in Portugal in 2017 [21], sugar concentration of most samples were found to be below to 8 g/100 mL which were less than those from 2008.

According to sugars contents of the samples declared on the Nutrition Facts labels, 20 out of 25 samples contain at least one of the ‘added (free) sugars’ of sucrose, glucose syrup, fructose syrup, and beet sugar. It was specified on the label of these 20 products that sucrose, fructose syrup, and glucose syrup were added to 20, 13, and 13 out of 20 samples, respectively. However, results obtained from HPLC showed that SD5, SD14, and SD24 did not contain sucrose sugar. This may be because consumers consider sucrose as less harmful than fructose and glucose sugar syrups in Türkiye. For this reason, the manufacturers misled consumers by stating that they added sucrose sugar even if they used glucose and fructose. SD1, SD2, SD8, SD9, SD11, and SD24

samples were labelled as no sugar added samples given in Table 1. According to Turkish food codex, there is no tax on beverages that do not contain added sugar. In this case, samples of SD1, SD2, SD8, SD9, SD11, and SD24 that are labelled as ‘no sugar added’ are not taxed. Furthermore, regardless of their sugar content SD22 and SD23 coke samples are taxed at 35% and other all samples are taxed at 10%. In this case, 19 out of 25 samples are taxed in Türkiye [4].

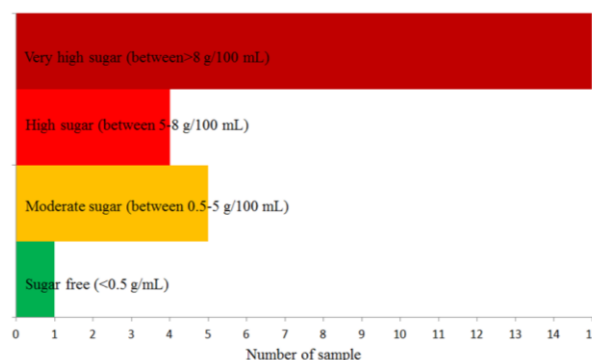


Figure 5. The classification of the samples according to their total sugar levels

Table 2. Sugar, HMF, pH, and titratable acidity values of the samples

Sample ID	Sugar Profile (g/100 mL)					HMF (mg/L)	pH	Titratable acidity (mEq/L)
	Fructose	Glucose	Sucrose	Maltose	Total			
SD1	2.74±0.02	2.72±0.05	3.48±0.04	ND	8.94	ND	3.41±0.01	14.20±0.35
SD2	3.06±0.01	3.16±0.02	2.77±0.02	ND	8.99	3.39±0.06	3.38±0.01	11.67±0.12
SD3	4.27±0.03	4.05±0.07	0.96±0.04	ND	9.28	6.00±0.08	3.83±0.01	5.73±0.12
SD4	2.55±0.02	2.84±0.04	8.46±0.04	ND	13.85	ND	2.78±0.01	6.67±0.12
SD5	5.82±0.04	5.18±0.09	ND	0.31±0.01	11.31	3.07±0.05	2.49±0.01	4.73±0.12
SD6	1.76±0.01	1.78±0.01	8.79±0.03	ND	12.33	0.65±0.01	2.60±0.01	5.80±0.20
SD7	5.23±0.24	3.86±0.14	0.50±0.01	ND	9.59	22.72±0.18	2.86±0.01	5.27±0.12
SD8	6.46±0.02	3.62±0.07	1.60±0.04	ND	11.68	8.13±0.06	3.61±0.01	4.13±0.12
SD9	5.62±0.02	6.31±0.05	ND	ND	11.93	16.82±0.44	2.93±0.01	22.07±0.12
SD10	2.09±0.02	2.91±0.11	6.82±0.2	ND	11.82	1.21±0.01	3.13±0.01	11.20±0.00
SD11	1.27±0.01	1.98±0.10	ND	ND	3.25	ND	4.24±0.01	5.13±0.12
SD12	3.39±0.02	3.59±0.15	0.60±0.02	ND	7.58	1.21±0.01	2.36±0.01	8.27±0.12
SD13	0.27±0.01	0.08±0.00	2.75±0.02	ND	3.10	ND	2.78±0.01	6.00±0.12
SD14	2.74±0.01	2.02±0.02	ND	ND	4.76	2.16±0.09	2.54±0.01	4.67±0.20
SD15	1.87±0.02	1.89±0.03	3.53±0.06	ND	7.29	ND	2.68±0.01	6.13±0.12
SD16	1.70±0.01	1.50±0.01	4.64±0.13	ND	7.84	0.80±0.03	3.19±0.01	3.67±0.12
SD17	1.41±0.04	1.37±0.05	2.24±0.05	ND	5.02	ND	3.15±0.01	3.27±0.12
SD18	1.43±0.02	0.57±0.02	2.40±0.02	ND	4.40	6.49±0.14	3.15±0.01	2.93±0.12
SD19	1.24±0.00	1.19±0.00	10.79±0.02	ND	13.22	ND	1.99±0.01	12.27±0.20
SD20	0.52±0.01	2.18±0.03	8.41±0.07	ND	11.11	2.59±0.01	2.97±0.01	12.33±0.12
SD21	0.71±0.01	0.89±0.04	9.16±0.02	ND	10.76	ND	2.36±0.01	5.80±0.23
SD22	2.43±0.01	2.58±0.02	6.04±0.07	ND	11.05	ND	1.96±0.01	4.93±0.12
SD23	4.55±0.00	5.06±0.04	1.21±0.01	ND	10.82	1.82±0.08	2.03±0.01	3.33±0.31
SD24	ND	ND	ND	0.23±0.01	0.23	27.76±0.08	6.34±0.01	0.73±0.12
SD25	1.02±0.00	0.98±0.08	7.63±0.08	ND	9.63	2.39±0.12	2.21±0.01	8.27±0.20
Average	2.57	2.49	3.71	0.02	8.79	4.29	3.00	7.17

ND: Not detected

3.2. HMF Analysis by HPLC-DAD

Generally, the main sources of HMF are sugars such as glucose and fructose. It occurs in the presence of acids at high temperature thermal processing and bad storage conditions during the Maillard reaction and caramelization. It is commonly found in sugar syrups

and thermally processed foods such as fruit juices, vinegars, baby food, honey and coffee [13]. Therefore, quantification of HMF analysis in soft drinks is very crucial and can be counted as one of the indispensable quality parameters. However there are legal limits set for some foods such as honey [22], and there is no limit established for the concentration of HMF in soft drinks.

The HMF concentrations of the samples were calculated by Agilent LC solutions software using peak areas of calibration curves. An example of HMF sample chromatogram is given in Figure 6.

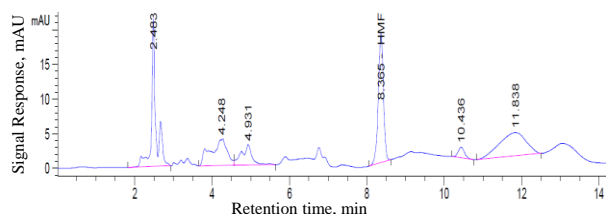


Figure 6. An example of a typical sample chromatogram for HMF component

HMF was not detected in 9 out of 25. The level of HMF content of the samples was found to be ranging from 0.65 to 27.76 mg/L and the mean value of the samples was detected as 4.17 mg/L. Although the lowest total sugar content (0.23 g/100 mL) and the highest pH (6.34) was identified in the iced coffee sample, the highest amount of HMF content was detected as 27.76 mg/L. This could be as a result of coffee roasting and exposing it to high temperature [23]. HMF content in sugar syrups is seen as major public health concern. The generation of HMF in syrups could be as a result of the heating process [24]. Thus, the addition of sugar syrups may cause an increase in the HMF content of the samples. HMF contents of SD7 (sugar-added apple juice) including sucrose / fructose-glucose syrup and SD8 (squeezed apple juice) no sugar-added samples were detected as 22.77 and 8.13 mg/L, respectively. The sugar-added apple juice (SD7) had almost 3 times more HMF than squeezed apple juice (SD8). In a previous study performed by Czerwonka et al. [25], HMF content in different beverages including apple juices, tomato juices, blackcurrant nectars, coke drinks, and citrus flavoured soft drinks was examined. HMF contents were found in a very wide range of values ranging from 0.23 mg/L to 110.75 mg/L. The highest and the lowest mean HMF contents were detected in apple juice samples and orange juice samples as 17.33 and 0.87 mg/L, respectively. Furthermore, HMF levels of roasted and instant coffee samples were also determined. The coffee samples were prepared from 10 g and 6 g of coffee beans and instant coffee assuming them as a cup of ready-to-drink coffee. The results show that HMF contents of the coffee samples were found to be relatively high compared to the drink samples. Mean HMF values of the roasted and instant coffee samples were determined at the amount of 347.6 and 3351 mg/L, respectively. It is stated that, the reason of instant coffee has about ten times higher than roasted coffee was due to instant coffee being subjected to heat treatment during production process.

3.3. pH and Titratable Acidity

The terms of pH and acidity are interrelated to each other representing acidity in aqueous media [26]. In general, soft drinks are naturally acidic [27]. Drinks that are not naturally acidic gain acidity with the addition of acidity regulators which are defined as acidulants. Acidulants as

an essential part of soft drink formulations are also added for the inhibition of microbial growth to extend the shelf-life of the products. Furthermore, they are added to balance the sweetness. Consequently, acidulants are giving them a distinctive taste [28, 29, 30]. According to labelling information, all the 'sugar-added' soft drink samples contain at least one acidulant. Citric acid, trisodium citrate, malic acid, concentrate lemon juice, sodium citrate, sodium bicarbonate, phosphoric acid, and ammonium sulfate (see Table 1) are used as acidulants in the samples. The soft drink samples that are free of added sugar do not contain acidulants except sample of SD24 (iced coffee). Among the acidulants, the most commonly used acidulant was citric acid. It was added in 13 out of 25 samples. Furthermore, concentrated lemon juice was used in 3 samples as a source of citric acid as well. This acidulant is commonly required in sucrose-based carbonated soft drinks due to its relatively low toxicity compared to other acidulants. In addition to that, it is also used for pH adjustment and stronger tartness because it has relatively high equivalent weight (70.05 EW) compared to other acidulants such as malic acid, phosphoric acid, and acetic acid [26, 31].

The pH values of the samples ranged between 1.96 and 6.34 and the mean value of the samples was found to be at the amount of 3.12. The lowest and the highest pH values are detected in SD22 coke sample and SD24 iced coffee sample as 6.34 and 1.96, respectively (see Table 2). Titratable acidities of the samples were detected as ranging between 0.8 and 14.80 mEq/kg and the mean values of the samples were found at the amount of 9.92 mEq/L. The highest and the lowest titratable acidity were found in the SD9 squeezed pomegranate juice sample and SD24 iced coffee sample at the amounts of 22.07 and 0.73 mEq, respectively (see Table 2).

Although acidulants and low pH provide advantages for the end product in the beverage industry mentioned above, they have negative effects on consumer's health. It has been reported in the study that soft drinks that have low pH and high acidity cause tooth erosion including enamel erosion and dental material corrosion [32, 33, 34]. Reddy et al. [28] classified the corrosive degree of soft drinks based on apatite solubility quoted from a study conducted by Larsen and Nyvad [32]. According to this classification soft drinks were classified based on their pH values as extremely erosive (pH<3), erosive (pH between 3 and 4), and minimally erosive pH ≥4 (see Figure 7).

Taking into account of this classification, most of the pH values of the samples were considered as extremely erosive. Among the samples, 14 out of 25 samples were found to be extremely erosive, 9 samples were detected as erosive, and 2 samples were in the class of minimally erosive (see Figure 8). Reddy et al. [28] collected 379 soft drink samples including juices, energy drinks, sodas, flavoured waters, iced teas for pH analysis. It was concluded that 39% of the samples were found extremely erosive, 54% of the samples were considered erosive and only 7% (25 out of 379) of the samples were classified as minimally erosive.

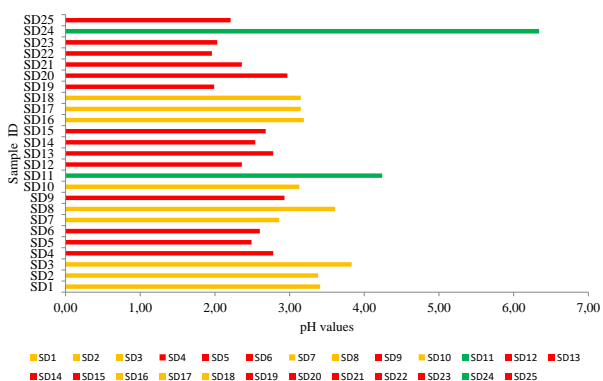


Figure 7. pH values and erosive capacities of the samples (Green, orange, and red bars indicate the erosive capacity of the drinks. Red is for extremely corrosive, orange is for erosive, and green is for minimally erosive)

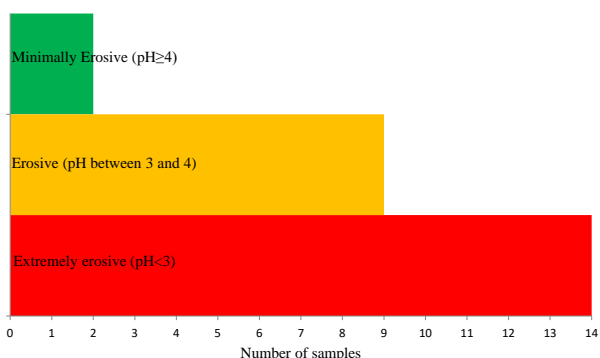


Figure 8. Relative erosivity of the samples (Green, orange, and red bars indicate the erosive capacity of the drinks. Red is for extremely corrosive, orange is for erosive, and green is for minimally erosive)

3.4. Pearson Correlation Matrix Among the Total Sugar, HMF, pH, and Acidity Values

Pearson correlation was conducted in the IBM SPSS Statistics software package program It was found that Pearson correlation coefficients (PCC) among the total sugar, HMF, pH, and acidity values show that there were negative correlations among the all parameters. The highest and the lowest PCC value were obtained between HMF – pH and acidity – HMF as -0.583 and -0.022, respectively. Furthermore, PCC values for total sugar – pH, total sugar - HMF, pH – acidity, and total sugar – acidity were found as -0.583, -0.245, -0.210 and -0.450, respectively (see Figure 9).

P	Total sugar	HMF	pH	Acidity
Total sugar	1			
HMF	-0.245	1		
pH	-0.583	-0.588	1	
Acidity	-0.45	-0.022	-0.21	1

Figure 9. The Pearson correlation coefficients between the parameters (in figure, P represents parameter)

4. CONCLUSIONS

While, among the sugars; fructose, glucose, sucrose and maltose, the most frequently found sugar was sucrose at the average level of 3.71 g/100 mL in the samples, maltose was found only in two samples and had an average of 0.02 g/100 mL. The most common sugars were fructose and glucose detected in 24 out of 25 samples. Most of the samples (15 out of 25) were to be in a ‘high sugar content’ class ranging between 8.94 and 13.22 g/100 mL. HMF was identified in 16 out of 25 samples ranging from 0.65 to 27.76 mg/L. pH values of the samples were found to be in very wide ranges changing between 1.96 and 6.34. According to erosive capacity, 14 out of 25 samples were considered to be in extremely erosive class (pH<3). The titratable acidity of the samples was found to be between 0.8 and 14.80 mEq/L. As a conclusion, most of the samples were found to be categorised in ‘high sugar content’ and ‘extremely erosive’ classes. From this perspective, it can be said that the soft drink samples could have negative impact on the consumer’s health.

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