DETERMINATION OF CONSTITUENTS AND ESTIMATION OF SUGAR AND TRACE ELEMNET IN SOFT DRINKS

AMINA SULTANA[1], ANILA ANWAR[*1], RUBINA PERVEEN[1], FARAH KISHWER[1], RABIA FARHEEN[1], AZHAR ALI[1] AND SHAFQAT HUSSAIN[2]

[1]Department of Chemistry, Federal Urdu University of Arts, Science and Technology, Gulshan-e-Iqbal, Karachi, Pakistan
[2]Department of Chemistry, Karakoram International University, Gilgit-15100, Gilgit, Biltistan, Pakistan,
Corresponding author's e-mail: khan.anila anwar@gmail.com

خلاصہ

سوفٹ ڈرنک اپنی مخصوص ذائقہ. پیاس بجھانے کی صلاحیت اور آسان قیمت خرید ہونے کی وجہ سے پوری دنیا میں عام استعمال ہوتی ہے۔ طلب رسد کی حد پوری کرنے کے لئے نا صرف کوالٹی بلکہ اس میں دوران پیداوار ممکنہ بھاری دھاتی عناصر کی آمیزش پر مصالحت کرلی جاتی ہے جو انسانی صحت لے لئے انتہائی مضر ہو سکتی ہے۔ لہذا اس تحقیقی مطالعہ میں کراچی کی مارکٹس میں دستیاب مختلف سوفٹ ڈرنکس (Marinda, 7up and Pepsi) کے طبعی خواص (conductivity, TDS and pH)، ان کے اجزاء ترکیبی کی کیفی تشریح، شکر اور بھاری دھاتوں (Fe and Ni) کی کمی تشریح کی گئی ہے۔ کیفی تشریح میں تمام نمونوں میں شکر اور کاربن ڈائی آکسایڈ کی موجودگی ظاہر ہوئی، فاسفیٹ صرف ایک نمونے (Pepsi) میں جبکہ الکوحل کسی بھی نمونے میں نہیں پایا گیا۔ Specific gravity کے طریقے سے شکر کی مقدار (7up, Pepsi and Marinda) میں بتدریج 40.841g, 41.605g and 48.791 g / 345mL معلوم ہوئی جو کہ نمونوں کے لیبل پر بتدریج درج 37 ,37.3 and 46.2 g / 345 mL کے لحاظ سے زیادہ تھی۔ آئرن کی تخمین بذریعہ Spectrophotometric Technique کی گئی۔ 7upاور Marinda میں آئرن لیول بتدریج $1.5x10^{-6}$ M and $1.0x10^{-6}$ M پایا گیا جو کہ RDA حد سے کم ہے۔ Pepsi کے نمونے ان دونوں دھاتوں سے پاک ثابت ہوئے۔ Gravimetric method سے Nickal کی تشریح کی گئی اور تینوں نمونوں میں غیر موجود پایا۔

Abstract

Soft drinks are used all over the world due to its affordability, characteristic taste, and thirst quenching potential. However, its demand may compromise on the quality and possible contamination of heavy metals during production which may risk to human health. This study was carried out on physical parameters (conductivity, TDS, and pH), qualitative evaluation of some constituents and quantitative analysis of sugar and presence of heavy metals (Fe and Ni) contaminants in three different samples of soft drink (7up, Pepsi, and Mirinda) available in Karachi local markets. The qualitative analysis revealed the presence of sugar (sucrose) and carbon dioxide in all samples of soft drinks and phosphate in one sample while alcohol was absent in all samples. The sugar level through specific gravity method was found to be 40.841g, 41.605g, and 48.791g/345mLin 7up, Pepsi, and Mirinda respectively, which are higher than mentioned on label of sample bottles (37.3, 37 and 46.2g/345mL respectively). By using spectrophotometric technique concentration level of Fe was found to be $1.5x10^{-6}$ M and $1.0x10^{-6}$ M in Marinda and 7up sample respectively that is low in comparison of RDA limit. Pepsi sample bottle was free from this metal. Nickel was determined gravimetrically and not found in all samples.

Key words: soft drinks, pH, sugar level, iron concentration level

Introduction

The main source of energy to all living beings are carbohydrates. These are produced by plant through the process of photosynthesis as mono, di and polysaccharides (Ponnadurai *et. al*., 2004, Kerem *et al*., 2014). Glucose (mono), sucrose (di) and starch (poly) saccharides are the most commonly used carbohydrates (Rambla *et al.,* 1998). Sucrose generally known as sugar, is a natural polymer of glucose and fructose (Solomons and Craig, 2004). Owning sweet taste it is used as a sweetener in many food and beverages (Hansson *et al*., 2001). The soft drink is dissolved carbondioxide, non-alcoholic, fizzy drink which is used in chilled condition (Bryndolfaswon,1982). Having peculiar flavor, unique taste, thirst quenching with refreshing ability and affordability, soft drinks are universally used by general public in traditional celebrations especially during summer (Chua and Teo, 2017).The soft drink charms populace due to its components like sugar (sweet. energizing), carbonated water (fizzy, tangy), acids and phosphates (sugar-water balance, pleasant) (Aloh *et al.,* 2015).A healthy person can digest 2-3 tea spoon sugar without adverse health effects (Ponnadurai *et al.,* 2004).

Nevertheless excessive intake may risk for diabetes, obesity, cardiac problems, high B.P. dental issues (Emily *et al*., 2010) and hypoglycemia (Efrain *et al*., 1995). However, high demand of soft drinks may compromise in quality and production processes. Therefore, it may be contaminated by heavy metals if using impure water, contaminated fruits and other ingredients during manufacturing and by metallic packaging (Barbara *et al*., 2015,Helen *et al*., 2011).If heavy metal sexceedin diet, may deposit in vital soft tissues, leads to cancer, tumor, bronchitis, loss of memory, dizziness and allergies (Szyczewski *et al*., 2009, Kasprzak *et al*., 2003).
As an excess of sugar and heavy metal sconsequences in health issues (Said. *et al*., 1987), it is therefore imperative to know what amount of sugar and metal contamination is present in soft drink that we usually consumed. Thus, here in this study we evaluated some constituents of soft drinks which are generally consumed in Pakistan by reported protocols, amount of sugar added was estimate during specific gravity method, spectrophotometry method was used for determination of possible contamination of heavy metal (Fe) and Ni was determined gravimetrically.

Table 1. Physical parameters of soft drinks

S.No.	Samples	conductance (μS)	TDS (mg/L)	pH	$[H^+]$ion in mole
1	7 up	914	546	3.18	$6.606x10^{-4}$
2	Pepsi	1608	1254	2.47	$3.3x10^{-3}$
3	Miranda	894	535	2.84	$1.445x10^{-3}$

TDS=Total dissolved solid

Table 2. Qualitative analysis of soft drinks.

S.No.	Samples	Test for sugar	Test for reducing sugar	Test for PO_4^{-3}	Test for alcohol	Test for CO_2
1	7up	+	+	_	_	+
2	Pepsi	+	+	+	_	+
3	Miranda	+	+	_	_	+

Key: (+)indicatespresent,(⁻) indicates absent

Table 3. Quantitative analysis of sugar in soft drinks

S.No.	Samples	Specific gravity of sugar	Percentage of sugar(%)	Amount of sugar(g)
1	7up	1.04582	11.4	40.841
2	Pepsi	1.04668	11.6	41.60
3	Miranda	1.05472	13.5	48.79

Table 4. Quantitative analysis of iron and nickel in soft drinks

S.No.	Sample	Concentration of iron [M]	Concentration of nickel [M]
1	7 up	1.0×10^{-6}	ND
2	Miranda	1.5×10^{-6}	ND
3	Pepsi	ND	ND

Key: ND = Not detected

Materials and Methods

Sample collection: All samples of soft drinks were collected from market retail stores in Karachi city of Pakistan.

Experimental: All chemicals purchased from Sigma - Aldrich Chemicals are of analytical grades. pH is measured by digital pH meter (3510, UK), conductivity and TDS (total dissolved solid) were measured by conducto meter (4510,UK). Specific gravity was determined using digital weighing balance (TP-214, UK) and double-beam spectrophotometer UV-Visible, 1800 Spectrophotometer Schimadzu Japan was used to quantify metal contamination.

Qualitative Analysis: The qualitative analysis were carried out at ambient conditions but quantification of sugar was done at 20^0C. All qualitative analysis was done according to the protocols reported in literature**.**

Determination of physical parameters: The conductance and total dissolved solids were determined by conducto meter. pH was found by pH meter and $[H^+]$ion concentration was calculated through this pH value.

Test for reducing sugar: The occurrence of reducing sugar was determined by means of Fehling's solution test (Nekrasov, 1978).For this 3 mL of the soft drink sample in a test tube was mixed with equal volume (2 mL) of both A and B Fehling's solutions. Heated for 10 min. in a water bath.The appearance of red-brown precipitation confirmed the existence of reducing-sugar.

Test for sugar: The existence of non-reducing sugar was determined after hydrolysis of sugar (Nekrasov, 1978). Add 0.5 mL of conc. HCl to 3mL of soft drink sample in a test tube. Heat the mixture in a water bath for 20 minutes. After cooling 1M NaOH was added till the solution test neutral on litmus paper. Add 1mLof Fehling solution A and 1mL of Fehling solution B reflux for 10 minutes. The appearance of comparatively boosting red precipitatesthan in test of reducing sugar indicates positivity for the presence of sugar (sucrose).

Test for phosphates: In a test tube 3 mL sample of soft drinks was mixed with 2 mL ammonium molybdate and 2 mLconcHNO3. The mixture was refluxed for about 10 mins. in a water bath. The phosphate ion was confirmed by appearance of canary yellow color ppt. (Engwa *et al*., 2015).

Test for alcohol: In a test tube3 mL of sample was mixed with equal volume (1mL) of each iodine, KI and aq NaOH. Boil the mixture in a water bath at 100^0C for 30 mins. No yellow colored precipitate were observed. Confirmed absence of alcohol in soft drinks (Engwa *et al*., 2015).

Test for carbon dioxide: Instantly after opening of sample bottle, 3 mL sample of soft drink was pouredinto lime water (2mL) taking in a test tube. The lime water changed from colorless to milky. It confirm edthe presence of dissolved carbon dioxide in the soft drink (Ahluwalia *et al*., 2005).

Determination of acid content: The pH of the sample was determined by a digital pH meter at room temperature (Engwa *et al*., 2015). For this 5mLsample was taken in a 25mL beaker. The pH meter electrode dipped in sample then turned on for measurement. The acid content in terms of hydrogen ion concentration was calculated by pH values using formula: $pH = -\log [H^+]$

Quantitative analysis of sugar: Amount of sugar in soft drinks was determined by specific gravity method. At first all samples of soft drinks were degassed. Weight of 100mL distilled water taken in 100mL volumetric flask was determined at exactly 20^oC over a digital weighing balance. Then 100mL degassed soft drink was taken in the same 100mL volumetric flask and weighed at same temperature over same balance. The specific gravity of the soft drink was then measured with reference water. The percent sugar at this value of specific gravity was found from the reported table of William Horwitz (1980).

Spectrophotometric determination of iron concentration: The concentration of iron in soft drinks was determined by the method of Gary (2004).In 250 mL volumetric flasks, prepare stock solution of iron. Then a series of standards was prepared from it in100mL volumetric flask. Add 1.0 mL of the hydroxyl ammonium chloride solution, 5.0 mL of the 1,10- phenanthroline solution and 8.0 mL of the sodium acetate solution as a buffer to produce the red color complex in each series. Allow all solutions at least 15 minutes to stand at room conditions then take absorbance at 510nm on spectrophotometer. Plot the absorbance against the wavelength then prepare a calibration curve to measure absorbance of unknown sample of iron.

Gravimetric estimation of nickel: Nickel was estimated according to the method Ziauddin *et al.,* (1999).Take 10mL of sample solution in a beaker then add 2 drops of concentrated HCL. Now add 10 mL DMG solution and 4 % ammonium hydroxide solution with continuous stirring till red precipitate obtained. These red ppt. were filtered through filter paper, washed with ammonical solution and weighed after drying in oven.

Results and Discussion

It is revealed from the above literature that sugar sweetness and thirst reducing aptitude of carbonated water are the most valid reasons for the excessive consumption of soft drinks all over the world. Therefore in the present research different constituents and amount of sugar along with trace element in the extremely used soft drinks 7up, Pepsi and Miranda have been determined. All the three samples of soft drinks were found acidic byshowing H^+ concentration between $1.445 \times 10^{-3}$to 6.606×10^{-4}g /L (Table-1). Even though low acid concentration is important in killing gastrointestinal bacteria but it may possibly cause teeth decaying (Emily *et al*., 2010).

Qualitative analysis determined the presence of both sucrose (non-reducing sugar) and glucose (reducing sugar) in all samples. These are the chief sweetening ingredients of any soft drink. Carbon dioxide which provides fizzy effect was present in all samples. Phosphorous is an important element of DNA and cell

membrane. It is essential for the growth of boney skeleton and teeth. Naturally exist as phosphates(Efrain *et al*., 1995). It was detected in only one sample of the soft drinks and thus could be advantageous for body growth. Alcohol was absent in all three samples of soft drink Table-2.

Table -3 shows the quantitative extent of sugar present in each bottle of soft drinks. The kind of sugar confirmed was sucrose (a non- reducing sugar).The reducing property of sucrose can be elicited only by acid hydrolysis. The sugar level was found to be 40.841, 41.6059 and 48.79g in 345mL of 7Up, Pepsi and Mirinda respectively. Miranda, however, contained the highest percentage of sugar level per 345mL (13.5%) while 7up contains the least percentage (11.4%) whereas Pepsi revealed 11.6% sugar per 345mL. The sugar level in this analysis was found in order of Mirinda > Pepsi >7up. The sugar level found is comparatively little higher than the values reported on the labels of the sample bottles.

Moreover, if impure water is used during the production process, there may possibly be contamination of heavy metals. Hence, dangerous to public health (Szyczewski *et al*., 2009).The concentration level of iron (Fe) was detected$1.5x10^{-6}$ M and $1.0x10^{-6}$ M in Mirinda and 7up samples respectively. The analyzed level of iron is comparatively low to the RDA limit and within the range of reported work. However, iron was absent in sample of Pepsi.In this study Ni was not determined in all three samples of soft drink Table -4.

Conclusion

The results concluded that the main sugar in mentioned samples soft drinks is sucrose which is a non-reducing sugar, its presence was confirmed after acid hydrolysis. Though the quantity of sugar in these soft drinks is comparatively slightly high than showed on the labels of the sample bottles (46.2g in Mirinda, 37g in Pepsi and 37g in 7up per 345mL), it must be generally reduced. Moreover, these soft drinks possesses pH ranging 2 to 3, are mostly highly acidic. Hence, soft drink consumption may generate a foremost public health problems that concern for high level of sugar, acid and heavy metal contamination. So, it is needed to monitor and control the quality of soft drinks in order to ensure safe intake and lesser public health risk.

References

Ahluwalia, V. K., Sunita, D. and Adarsh, G. (2005). College Practical Chemistry, Universities Press Pvt. Ltd., *Hyderabad*, India. p.12.

Aloh,G.S., Obeagu, E. I., Odo, C. E., Kanu, S.N., Okpara, k. E. and Nka, J. S. (2015).Estimation of sugar in soft drinks. *World Journal of Pharmacy and Pharmaceutical Sciences*, 4 (3): 112-125.

Barbara, B. A., Francisco, Daniel, M. B. and Ricardo, J. C.(2015). Determination of metals in soft drinks packed in different materials by ETAAS. *Food Chemistry* 185 : 488–494

Bryndolfaswon, (1982) "Soft drinks" Encyclopedia of Science and technology. Edited by Peterson J. Wlc-Graw Hill Press, New York.

Bueche, F. (1984). Principles of Physics. 4thEdt.,McGraw-Hill Book Company, Singapore, p. 208-209.

Chua, S. L. and Teo, S.S. (2017). Analysis of saccharin and benzoic acid in regular and diet Cola-flavored carbonated soft drinks. *International Journal of Food Science and Nutrition*, 2 (4): 58-64

Efrain, M. R., Fernando, G. R., Martha, R. M., Gloria, L.B., Ramon, P. and Dante, A. (1995). Consumption of soft drinks with phosphoric acid as a risk factor for the development of hypocalcemia in children: A case-control study. *The Journal of' Pediatrics*, 126 (6): 940-942

Emily, E. V., Jaimie, N. D. and Michael, I. G. (2010). Sugar Content of Popular Sweetened Beverages Based on Objective Laboratory Analysis: Focus on Fructose Content. Obesity, doi:10.1038/oby.2010.255

Engwa, A. G., Ihekwoaba, C. J., Ilo, U. S., Unaegbu, M., Ayuk, L. E. and Osuji, A. G. (2015).Determination of some soft drink constituents and contamination by some heavy metals in Nigeria. Toxicology Reports, 2: 384–390.

Gary, D. C. (2004). Analytical Chemistry. 6th Edition, Jhon Wiley and Sons, New Yark. P.765-766.

Hansson, A., Andersson, J. and Leufvean, A. (2001). The efect of sugars and pectin on flavour release from a soft drink-related model system. *Food Chemistry* 72 : 363-368

Helen, C., Rezende, Clesia C., Nascentes, Nivia, M.M. and Coelho. (2011). Cloud point extraction for determination of cadmium in soft drinks by thermospray flame furnace atomic absorption spectrometry. *Microchemical Journal*, 97 :118–121

Kasprzak, K. S., Sunderman, F. W. and Salnikow, K. (2003). Nickel carcinogenesis. Mutat. Res, 533: 67-97.

Kerem, I., Ismail, H. B. and Ali, T. (2014). Rapid analysis of glucose, fructose and sucrose contents of commercial soft drinks using Raman spectroscopy. *Food Control*, xxx: 1-6.

Nekrasov, V.V. (1978). Practical Organic Chemistry A Basic Course, p.208-219, Mir Publisher, Moscow, *USSR*.

Ponnadurai, R., Sabina, J. L., Philipe, R., Frederic, C., Heena, S. and Ashwini, S.(2004) Quantification of Sugars in Soft Drinks and Fruit Juices by Density, Refractometry, Infrared Spectroscopy and Statistical Methods. S. *Afr. J. Chem*., 57: 24–27

Rambla, F.J., Garrigues, S., Ferrer, N. and Guardia, M. (1998). Simple partial least squares–attenuated total reflectance Fourier transform infrared spectrometric method for the determination of sugars in fruit juices and soft drinks using aqueous standards. Analyst, 123: 277–281

Robert, R., David, H. and Kenneth, S.K. (2002). Physics. Vol.1, 5thEdt.,Jhon Wiley and Sons, New Yark. p. 332-336.

Said, M., Rahman, M.A. and Dsilva, A. L. (1987). Elements in Health and Disease .Hamdard University Press Medinat al Hikmat Bund Murad khan. And Printers, Karachi, Pakistan, 122:124-125.

Solomons, T.W.G. and Craig, B. F. (2004). Organic Chemistry, 8th edition, John Wiley & Sons (Asia) Pte. Ltd., Singapore, P. 1102-1103.

Szyczewski, P., Siepak, J., Niedzielski, P. and Sobczyński, T. (2009). Research on Heavy Metals in Poland. Polish J. of Environ. Stud. 18: 755-768.

William, H. (1980). Official Methods of Analysis of the Association of Official. 13th edition, Association of Official *Analytical Chemist*. Washington DC.

Ziauddin, M., Arshad, H.S. and Arshia, S. (1999).Practical Chemistry: Inorganic and General, 5th edition, *FUUAST* Press, Karachi, Pakistan, P. 68-70.