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Trace analysis of environmental endocrine disrupting contaminant bisphenol A in canned, glass and polyethylene terephthalate plastic carbonated beverages of diverse flavors and origin

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Abstract

Bisphenol A (BPA) is a hazardous contaminant demonstrating endocrine disrupting properties, and assumed to be involved in the pathogenesis of various cancer diseases for instance prostate, lung and breast cancer. The objective of the present study was to estimate the BPA amounts in carbonated beverages from the Saudi Arabian market for the first time using an authenticated technique based on solid-phase extraction and ultra-performance liquid chromatography-tandem mass spectrometry. A total of thirty-four carbonated beverages of different flavors, origin and packaging materials were studied. The beverage production periods were from February 2018 to July 2018 containing volume (250-1000 mL), packaging materials were of canned, glass and polyethylene terephthalate (PET) plastic. BPA amounts in canned (0.64-11.41 μg/L), glass bottled (1.92-29.56 μg/L) and PET plastic bottled (0.37-21.83 μg/L) were obtained with recovery (97.64-99.96%). Relatively, glass bottled has offered higher amounts of BPA compared to PET plastic bottled and canned samples. The unforeseen presence of BPA especially in glass bottled emphasizes the ubiquity of such compound beside the food fabrication chain, far off to the food packaging materials. Thus, a further knowledge on BPA amounts in glass bottled samples, in addition to threat assessment studies, is essential to defend human health.

Keywords: bisphenol A; carbonated beverages; canned; glass bottled; polyethylene terephthalate plastic bottled.

Practical Application: Authenticity and traceability of BPA in carbonated beverages using SPE and UPLC-MS/MS.

1 Introduction

Bisphenol A (BPA) is one of the most expansively established resources in the routine application nowadays and the yields obtained from BPA constitutes the health threats to human being (Srivastava et al., 2015). BPA is an essential element of polycarbonate plastics, which is an insubstantial, great enactment plastic with limitless useful applications (Tzatzarakis et al., 2017; Thomson & Grounds, 2005; Dreolin et al., 2019; Goodson et al., 2002). BPA is also a vital component of epoxy resins, which are applied as defensive coverings on metallic cans to preserve the grade of canned drinks and foods (Tzatzarakis et al., 2017; Thomson & Grounds, 2005). In addition, polycarbonate plastics including epoxy resins used in an extensive variety of goods such as plastic bottles, dental fillings, food containers, thermal paper and optical discs (Tzatzarakis et al., 2017; Thomson, & Grounds 2005; Dreolin et al., 2019; Goodson et al., 2002). As a result, the exposure of BPA to human is ubiquitous and has been identified in many individuals (Calafat et al., 2005). BPA has been assumed to be a generative toxin at higher amounts, with adversative impacts on animal's mammary glands and kidney (European Food Safety Authority, 2015). Recently, many studies have revealed the effects of BPA on reproductive system, thyroid function, nervous system, respiratory system, cardiovascular disorders, obesity and cancer diseases (Shi et al., 2019; Kwon et al., 2020; Yau et al., 2015; Ma et al., 2019). Besides, rising information have advocated that there is a link between BPA amounts and the growth of tumors comprising lung, prostate and breast cancer (Zhang et al., 2014; Tse et al., 2017; Ben-Jonathan, 2019). Because of BPA lipophilic properties, it possibly be stored in adipose tissue which could consequently be an appropriate matrix to evaluate the long-term exposure to BPA and associations with the tumorigenesis of such cancer diseases. After assessing the obtainable scientific data, the European Commission directive 2004/19/EC has sets the BPA migration limit (0.6 μg/g) in material envisioned to come into contact with drink and food, and the acceptable daily consumption limit is 0.05 μg BPA/g body weight/day (European Food Safety Authority, 2006; European Union, 2004). Soon after, the EC directive 2011/8/EU forbidden the usage of BPA in baby bottles and limit the BPA migration from coatings or used for the preparation of materials (European Commission, 2011). Recently (2018), a new EC directive 2018/213/EU has been implemented setting a further preventive of BPA migration limit with no BPA migration from applied materials coatings or varnishes, and any objects precisely intended to newborns and young teenagers' maximum of three years old, is only allowable (European Union, 2018; European Commission, 2018). The Environmental Protection Agency (EPA) has also established a reference dose of 50 μg/kg bw/day (Environmental Protection Agency, 1993).

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As a result of these BPA limits, industrial establishments are progressively substituting BPA by means of other equivalent compounds (Pelch et al., 2017).

Because of BPA extensive and recurrent exposure, the big distresses relating to the BPA application drove a number of regimes to impose restrictions on BPA usage and leading to the desertion of BPA use by the companies. Taking into consideration, the Canada turn out to be the first nation to categorize BPA as a lethal compound and barred the use of BPA in baby bottles (Modern Testing Services, 2013). In view of the most earlier epidemiological investigations surveyed the cross-sectional links between determined urinary BPA amount and health consequences, an arbitraries mediation test can offer a superior level of indication and may explain the health influence of BPA more evidently. BPA could come into individuals via skin, respirational and gastrointestinal tracts, of which the gastrointestinal tract is known to be the main source of BPA absorption (Geens et al., 2012; Ye et al., 2012). The BPA could be identified in many human biological fluids for instance urine, blood and milk (Vom Saal & Welshons, 2014; Teeguarden et al., 2016).

Carbonated beverages are a common element of the present dietary way of life globally, and Saudi Arabia ranked top position in the consumption of carbonated beverages among Arab nations. In Saudi Arabia, nearly 56.3% and 17.1% youngsters were allegedly consuming carbonated beverages weekly and daily basis, respectively (Alsubaie, 2017).

To our knowledge, presently, there are no measureable summary assessment information available relating to the occurrence of BPA in carbonated beverages from the Saudi Arabian market. Thus, it was the objective of present study to estimate the BPA amounts in carbonated beverages of various flavors, origin and packaging materials using an authenticated technique based on advanced solid-phase extraction (SPE) and ultra-performance liquid chromatography-tandem mass spectrometry (UPLC-MS/MS).

2 Materials and methods

2.1 Chemicals and reagents

Bisphenol A (purity ≥99%) and bisphenol A-d_{16} 98 atom %D (BPA-d_{16}, internal standard, IS) were obtained from Merck (Darmstadt, Germany). The chemical structure of the BPA and BPA-d_{16} has been illustrated in Figure 1. LC-MS grade acetonitrile, methanol and dichloromethane, ethyl acetate, ammonia solution (25%) and formic acid (reagent grade, ≥95%) were purchased from Sigma-Aldrich (Missouri, USA). Sodium hydroxide was achieved from BDH Laboratory Supplies (Poole, UK).

Individual stock solution of BPA and BPA-d_{16} (IS) at concentration of 100 µg/mL were prepared in methanol and used for dilution calibration and standard addition solutions. For calibration curve, six individual solutions at levels of 0.2, 1, 5, 15, 35, 50 µg/L (w/v) were prepared. BPA-d_{16} (IS) was prepared at concentration of 0.2 µg/mL, and added to each calibration solutions and samples during analysis. All samples including stock and calibration solutions were refrigerated at 4 °C in amber glass vials.

The ultrapure (Milli-Q) water was acquired from Millipore Corporation Advantage A 10 water purification system (Bedford, USA). The vacuum manifolds for instance Visiprep and Visidry from Supelco (Gland, Switzerland) were applied for the extraction and evaporation of samples, respectively. The pH of the sample was carried out by pH meter (Thermo Scientific, Singapore). The mixing of the sample extracts was carried out by a vortex mixer (Stuart™ model SA8, Fisher Scientific, Göteborg, Sweden). The samples and standard solutions were filtered using mini centrifuge (Cole Parmer, Seoul, South Korea) and ultra-free centrifugal polyvinylidene fluoride (PVDF, 0.45 µm) filters (Millipore Corporation, Billerica, USA).

Extraction cartridges Bond Elut C_{18} (500 mg, 3 mL) from Agilent Technologies (Santa Clara, USA) were achieved and used for SPE purposes. The extraction Extrelut NT20 empty column and stopcocks were purchased from Merck (Darmstadt, Germany) and Varian (Harbor City, USA), respectively.

2.2 Sample and sample extraction

A total of thirty-four carbonated beverages of different flavors, origin and packaging materials were obtained from local markets based in Riyadh, Saudi Arabia. The beverage production periods were from February 2018 to July 2018 containing a volume of 250 mL to1000 mL, packaging materials were of can, glass and PET plastic. After purchasing, the samples were immediately refrigerated to evade any infective contamination. Table 1 displays the flavors, origin and packaging materials of studied samples.

The beverage samples preparation was carried out in contemplation with care processes to reduce any sample contamination. Glassware including other sample preparation materials were carefully washed with ultra-pure water followed by methanol to avoid any BPA contamination. To facilitate the extraction of BPA from beverage samples, the successive SPE procedure has been used. Primarily, the beverage samples

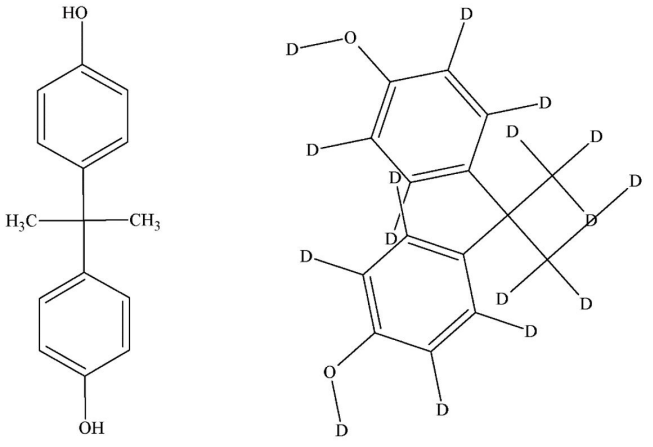

Figure 1. Chemical structure of the studied BPA and BPA-d_{16} (IS).

Table 1. BPA amounts and recovery values obtained in canned, glass bottled and PET plastic carbonated beverages.

Beverages	Flavor	Origin	Volume (mL)	Production date	Packaging material	Amounts of BPA before addition, (µg/L) ± sd	Amounts of added BPA, (µg/L)	Amounts of BPA after addition, (µg/L) ± sd	Recovery (%)
1B	Apple	KSA	250	Jun., 2018	Can	1.31 ± 0.01	2.00	3.28 ± 0.02	98.98
2B	Malt	KSA	250	Jun., 2018	Can	4.92 ± 0.03	5.00	9.92 ± 0.06	99.96
3B	Strawberry	KSA	250	Jun., 2018	Can	11.41 ± 0.06	11.00	22.00 ± 0.31	99.52
4B	Pomegranate	KSA	250	Jun., 2018	Can	7.09 ± 0.04	7.00	13.98 ± 0.15	99.21
5B	Apple	UAE	330	May, 2018	Glass	5.32 ± 0.03	5.00	10.20 ±0.06	98.86
6B	Malt	UAE	330	Mar., 2018	Glass	4.59 ± 0.03	5.00	9.50 ± 0.06	99.04
7B	Strawberry	KSA	330	Mar., 2018	Glass	3.59 ± 0.02	4.00	7.45 ± 0.05	98.15
8B	Pomegranate	UAE	330	Mar., 2018	Glass	3.28 ± 0.02	3.00	6.20 ± 0.05	98.72
9B	-	KSA	250	Feb., 2018	Glass	23.19 ± 0.32	23.00	46.00 ± 0.86	99.59
10B	Malt	Portugal	330	Apr., 2018	Glass	2.54 ± 0.01	3.00	5.48 ± 0.03	98.92
11B	-	KSA	250	Apr., 2018	Glass	4.03 ± 0.03	4.00	8.00 ± 0.04	99.62
12B	-	KSA	250	Mar., 2018	Glass	3.98 ± 0.03	4.00	7.85 ± 0.04	98.40
13B	Orange	KSA	250	Jul., 2018	Glass	18.18 ± 0.20	18.00	36.00 ± 0.67	99.51
14B	-	KSA	250	Apr., 2018	Glass	12.74 ± 0.14	13.00	25.30 ± 0.38	98.29
15B	Lemon	KSA	300	Apr., 2018	Glass	5.20 ± 0.04	5.00	10.00 ± 0.07	98.01
16B	Orange	KSA	330	Mar., 2018	Can	10.65 ± 0.06	11.00	21.30 ± 0.28	98.37
17B	-	KSA	330	Mar., 2018	Can	1.83 ± 0.01	2.00	3.78 ± 0.02	98.79
18B	-	KSA	330	Apr., 2018	Can	3.64 ± 0.02	4.00	7.46 ± 0.04	97.64
19B	Malt	KSA	500	Apr. 2018	Can	0.64 ± 0.001	0.60	1.22 ± 0.01	98.29
20B	-	KSA	355	Jun., 2018	Can	15.27 ± 0.09	15.00	30.00 ± 0.41	99.11
21B	-	KSA	355	Jun., 2018	Can	3.14 ± 0.02	3.00	6.07 ± 0.03	98.93
22B	Lemon	KSA	355	Apr., 2018	Can	2.66 ± 0.02	3.00	5.58 ± 0.05	98.51
23B	-	KSA	355	Jun., 2018	Can	3.80 ± 0.03	4.00	7.75 ± 0.06	99.36
24B	-	KSA	300	Mar., 2018	Glass	29.56 ± 0.45	30.00	59.20 ± 0.87	99.48
25B	Strawberry	KSA	330	May, 2018	Can	1.67 ± 0.01	2.00	3.63 ± 0.02	98.99
26B	Strawberry	KSA	250	Jun., 2018	Glass	1.92 ± 0.01	2.00	3.86 ± 0.02	98.57
27B	-	KSA	400	Jun., 2018	PET	5.81 ± 0.03	6.00	11.75 ± 0.06	99.46
28B	Orange	KSA	1000	May, 2018	PET	8.54 ± 0.04	9.00	17.40 ± 0.21	99.21
29B	-	KSA	1000	Jun., 2018	PET	5.90 ± 0.03	6.00	11.86 ± 0.06	99.66
30B	Strawberry	KSA	400	Jun., 2018	PET	1.08 ± 0.01	1.00	2.06 ± 0.02	99.05
31B	-	KSA	500	May, 2018	PET	9.64 ± 0.07	10.00	19.50 ± 0.25	99.28
32B	Orange	KSA	250	Feb., 2018	PET	11.35 ± 0.09	11.00	22.00 ± 0.31	99.33
33B	Lemon	KSA	1000	Jun., 2018	PET	0.37 ± 0.001	0.40	0.76 ± 0.001	98.97
34B	-	KSA	500	May, 2018	PET	21.83 ± 0.34	22.00	43.30 ± 0.64	98.83

sd = standard deviation (n = 3); can = tin packaging material; PET = polyethylene terephthalate; KSA = Kingdom of Saudi Arabia; UAE = United Arab Emirates; -described as original flavor.

were removed outside from the refrigerator, once the samples attained the room temperature, 10 mL of sample was taken out and put in a glass tube followed by sonication for 20 min to remove the gas from the sample. Afterward, the sonicated sample was placed into an empty Extrelut NT20 extraction column of 60 mL volume coupled to Bond Elut C_{18} (500 mg, 3 mL) cartridge, which was formerly preconditioned with 5 mL of ultra-pure water and 5 mL of methanol. Then, the beverage samples were eluted by means of SPE cartridge (Bond Elut C_{18}) at controlled flow rate of nearly 1 mL/min. After the complete elution of samples, a mixture of methanol and ultra-pure water (15 mL, 50:50, v/v) has been passed through the cartridge in order to remove the unwanted matrices, cartridge was drying for 10 min by vacuum manifold Visidry. Elution of the BPA from the cartridge was carried out by a mixture of ammonia and methanol (10 mL, 2:98, v/v). The sample extract was dried by mean of N_2 gas (purity, 99.99%). Finally, the dried sample was reconstituted with 1 mL mixture of ultra-pure water and methanol (50:50, v/v) containing BPA-d_{16} (IS) at concentration of 0.5 µg/L. The sample was vortex for 1 min followed by filtration through PVDF syringe filter (0.22 µm). The sample was collected in chromatographic vials (1.5 mL) and analyzed by UPLC-MS/MS system. The flow chart of the sample preparation method has been demonstrated in Figure 2.

In order to decrease the effect of matrix that impede with the compound signal identification, a precise standard addition quantification method was applied to measure the amounts of BPA in beverage samples. The standard addition procedure was performed using two unspiked samples at zero concentrations and four spiked samples at concentrations 0.5 µg/L, 1.0 µg/L,

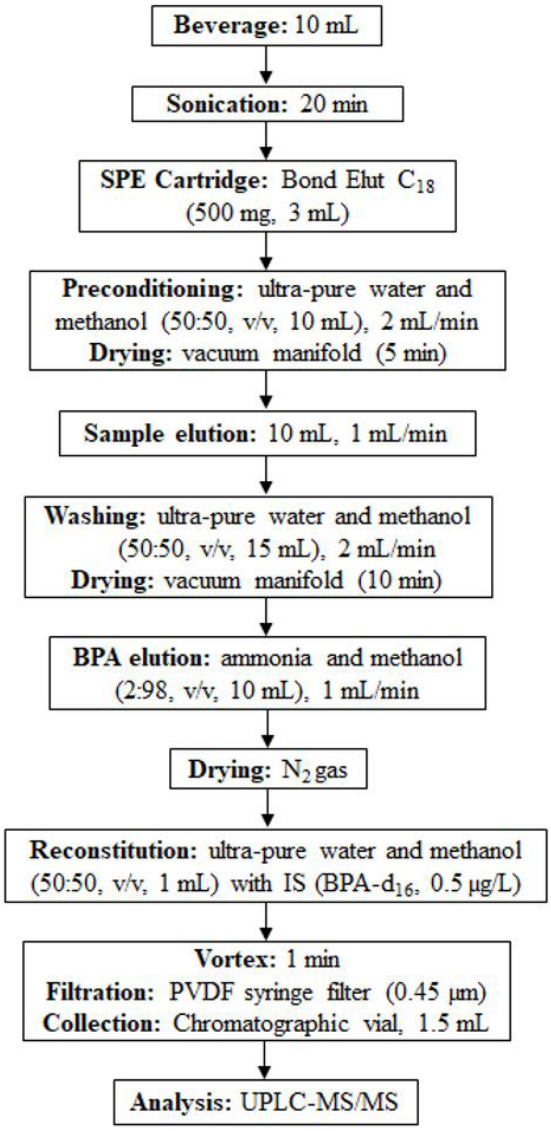

Figure 2. Flow chart of the sample preparation using SPE method.

10 µg/L and 50 µg/L concentrations. The beverage samples were analyzed in triplicates (n = 3). The BPA recovery was measured by means of the achieved linear regression slope between the added and found BPA amounts. In order to maintain the sensitivity of the UPLC-MS/MS instrument, blank and quality control samples were injected after each analyzed samples. Statistical data determination of the analyzed compound was carried out by means of the analysis of variance (ANOVA), a statistical method used to variances calculation between two or more means of the obtained concentrations.

2.3 Liquid chromatography

Ultra-performance liquid chromatographic (UPLC) system (Waters, Milford, USA) with solvent binary pump, column oven, auto sampler and vacuum degasser was coupled with a triple quadrupole mass spectrometer (MS/MS) equipped with electrospray ionization (ESI) source (Waters, Micromass, Milford, USA). The BPA analysis was carried out on Acquity BEH C_{18} reversed phase column 2.1 mm × 50 mm, particle size 1.7 µm (Waters, Milford, USA) with ambient column temperature. To retain the column efficiency and safety a guard column VanGuard™ BEH C_{18} with particle size 1.7 µm (Waters, Milford, USA) was applied during analysis. The elution of mobile phase was isocratic mode and comprises ultra-pure water (20%) and methanol (80%). The mobile phase flow rate was 200 µL/min. The injection volume of the sample was 5 µL with total analysis time of 2 min. The column was washed out every tem samples by a mixture of methanol and ultra-pure water, (50:50, v/v).

2.4 Mass spectrometry

The mass spectrometric acquisition was carried out on Waters TQD triple quadrupole mass spectrometer (MS/MS) fitted out with an ESI interface worked in negative ion mode and data was acquired in Multiple Reaction Monitoring (MRM) mode. The MS/MS ion transmission and their fragmentation patterns have been investigated via infusing a standard mixture of BPA and BPA-d_{16} (IS) at concentration of 10 µg/mL. BPA and BPA-d_{16} have been investigated as a precursor ion $[M–H]^-$ at *m/z*: 227.32 and *m/z* 241.20, respectively, the corresponding collision energy and cone voltage were optimized for higher intensity. The two product ions of BPA (*m/z*, 133.18 and *m/z*, 212.12) and BPA-d_{16} (*m/z*, 222.23 and *m/z*, 142.34) were obtained, the ions were applied for quantitative and qualitative analysis. The MS method was monitored using specific conditions for instance, ion source temperature (120 °C), desolvation temperature (350 °C), capillary voltage, (3.5 kV), cone voltage (25 V), desolvation gas (600 L/h) and cone gas (60 L/h). Nitrogen gas was used as cone gas generated from Peak Scientific (NM30LA) nitrogen generator (Inchinann, United Kingdom). Collision gas was used as argon achieved from Speciality Gas Centre (Jeddah, Saudi Arabia). Both gases were of high purity. The system vacuum was provided by a Oerlikon rotary pump (SOGEVAC SV40BI) (Paris, France). The most favorable collision energy, cone voltage and typical ions of BPA and BPA-d_{16} have been illustrated in Table 2. BPA data information was acquired by Waters MassLynx V4.1 software (Milford, USA).

3 Results and discussion

In order to authenticate the outcomes obtained from the analyzed beverage samples, primarily, the UPLC-MS/MS system has been optimized in terms of quality conditions for instance calibration correlation coefficient (R^2), detection limit (DL), quantification limit (QL) and precision. The obtained method quality conditions were R^2 (>0.9991), DL (0.02 µg/L), QL (0.04 µg/L) and precision (<2%) in terms of relative standard deviation (RSD%). In the current study, enhanced BPA DL and QL values were found as compared with the values (DL = 0.18 µg/L and QL = 0.54 µg/L) achieved in the previous study, analyzing carbonated beverages samples (Fasano et al., 2015). These values were found to be acceptable, and could be applied for the analysis of BPA in such type of beverage samples. The quality conditions of the applied method have been presented in Table 3. A total of thirty-four carbonated beverages of different flavors, origin and packaging materials were analyzed. The BPA amounts obtained in the studied beverages samples have been demonstrated in

Table 1. BPA were determined in each type of samples ranged from 0.37 µg/L to 29.56 µg/L. The amounts of BPA varied from 0.64 to 11.41 µg/L in canned, 1.92 to 29.56 µg/L in glass bottled and 0.37 to 21.83 µg/L in PET plastic bottled beverage samples. The higher amounts of BPA have been identified in glass bottled samples (29.56 µg/L) compared to PET plastic bottled (21.83 µg/L) and canned (11.41 µg/L) beverage samples. The recovery was obtained for canned, glass bottled and PET plastic bottled beverage samples ranged from 97.46 to 99.96%, 98.01 to 99.59% and 98.83 to 99.66%, respectively. The UPLC-MS/MS chromatogram of BPA obtained in beverage sample 13B has been illustrated in Figure 3.

Table 2. MRM mode used with MS/MS instrument[a] for the quantitative and qualitative BPA determination.

Analyte	Precursor ion, (*m/z*)	Quantification		Confirmation[b]	
		Product ion, (*m/z*)	Collision energy, (eV)	Product ion, (*m/z*)	Collision energy, (eV)
Bisphenol A	227.32	133.18	32	212.12	30
Bisphenol A-d_{16}	241.20	222.23	45	142.34	34

[a]Dwell time, 0.025 s; [b]ion intensity, >10%.

Table 3. Quality conditions of the applied UPLC-MS/MS method.

Analyte	Conc. Levels (µg/L)	R^2	DL (µg/L)	QL (µg/L)	Precision (RSD%)	
					run-to-run	day-to-day
Bisphenol A	0.2–50	0.9991	0.02	0.07	1.36	2.87

Conc. = concentration; R^2 = Correlation coefficient; DL = detection limit; QL = quantification limit; RSD = relative standard deviation.

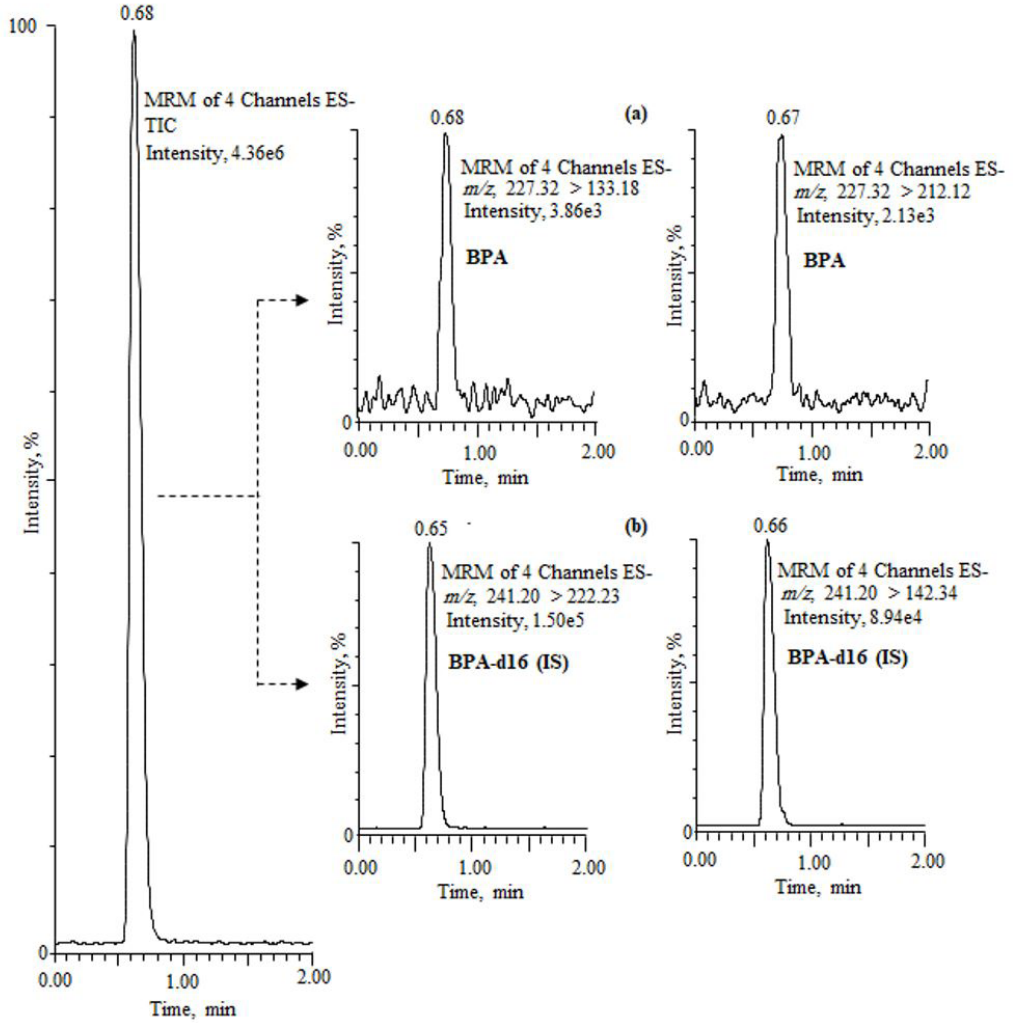

Figure 3. UPLC-MS/MS chromatogram of BPA (a) and BPA-d_{16} (b) obtained in beverage sample (13B).

In recent times, a rising figures of research apportioned with the presence of BPA especially in drinks and food products, as it contributes many chronic diseases (Srivastava et al., 2015; Rochester, 2013). The highest prospective for BPA adulteration of preserved drinks and food products is expected to be the migration from the inner coating of the container to the drinks and foods (González et al., 2019). In recent study, the authors have identified the BPA levels higher in solid type of preserved foods than in the liquid type of foods, even the BPA levels were much higher in preserved foods than in canned beverages (Geens et al., 2010). The BPA amounts in analyzed samples were compared with the recently BPA threshold established by the European Commission (50 µg/kg), and none of the studied samples were exceeded such limits (European Union, 2018; European Commission, 2018). Nevertheless, in some samples (9B, 24B and 34B) the maximum concentrations were obtained between 21.82 µg/L and 29.56 µg/L. In the current study, the BPA levels (0.64-15.27 µg/L) achieved in canned beverage samples (1B-3B, 16B-23B and 25B) were found to be in good agreements than the values (DL-1.26 µg/L) (Regueiro & Wenzl, 2015), 0.032-4.5 µg/L (Cao et al., 2009), <QL–8.10 (Geens et al., 2010) obtained in the earlier studies. Nevertheless, in canned food samples, the amounts of BPA were obtained at higher levels ranged from <0.17 to 88.66 µg/kg (González et al., 2019), 21.86 to 1858.71 µg/kg (Sungur et al., 2014) 0-842 µg/kg (Sajiki et al., 2007) as compared with the BPA detected in canned beverages, even exceeded the recommended BPA limit by the European Commission (50 µg/kg) (European Union, 2018; European Commission, 2018). From the comparative outcomes, the canned beverage samples contained lower amounts of BPA than the canned food samples, and canned beverages could be regarded as more safer than the canned foods. In glass bottled beverage samples (5B-15B, 24B and 26B), the BPA amounts were identified at concentrations from 1.92 to 29.56 µg/L. Comparatively, these values were found at higher concentrations than the canned beverage samples. The unexpected presence of BPA in glass bottled beverage samples emphasizes the ubiquity of such compound beside the food fabrication chain, far off to the food packaging materials. Nevertheless, Russo et al. (2019) have illustrated in the review literature that the BPA cannot be identified in glass bottled beverages but could only be identified in canned and plastic packed products due to the application of bisphenol A diglycidyl ether in the inner coating of beverage cans and therefore passes in the fabrication of epoxy phenolic resins (Russo et al., 2019). This information is completely contradictory in comparison with our results even BPA has been identified at higher concentrations. In the similar study, Tzatzarakis et al. (2017) have analyzed one glass bottled beverage sample and the BPA amount was detected at 1.1 µg/L. This value is consistent with the outcomes obtained in current analyzed samples (10B and 26B). Apart from this, a large number of glass bottled food samples have been studied earlier, and the BPA amounts were detected at different concentrations for instance 19.22-399.21 µg/kg (Sungur et al., 2014) and 0.27-1.28 µg/kg (Geens et al., 2010). Few studies relating to the presence of BPA in glass bottled beverage are available, however, more studies are needed to the verify the mechanism of BPA formation in such type of samples, even high BPA concentrations were determined but lower than the consumption limit suggested by the European Commission (European Union, 2018; European Commission, 2018). Relating to the BPA occurrence in PET plastic bottled beverage samples, (27B-34B), BPA were found at concentrations from 0.37 to 21.83 µg/L. The lower BPA value was found in 33B however, the sample 34B produced higher amounts of BPA but lower than the consumption limit suggested by the European Commission (European Union, 2018; European Commission, 2018). Tzatzarakis et al. (2017) have studied the BPA contents in plastic bottled beverage and identified the BPA amounts from <LOD-10.2 µg/L. These values were found to be more consistent with the values (except sample 34B) obtained in the current study. From the previously published literature sources, limited data are present relating to the occurrence of BPA in PET plastic bottled beverage samples. Nonetheless, a lots of data available on BPA occurrence in food products. Bemrah et al. (2014) have investigated canned and non-canned PET food products, and found the BPA levels from 0.045-4.510 µg/kg and 0.105-28.370 µg/kg, respectively. Chen et al. (2016) have studied the PET, plastic honey where the BPA was identified at concentrations 0.364-302 µg/kg. Cunha et al. (2011) have identified the presence of BPA in powdered infant formula where detected the amounts between 0.23 µg/L and 0.40 µg/L. These values obtained in food products were found to in good agreement with the values obtained PET plastic bottled beverage samples. However, the packaged food products were not investigated in the current study. The outcomes from the present and previous studies demonstrate the great variation in BPA levels and identified in almost each type of analyzed samples. The levels of BPA in our analyzed samples were identified at levels lower than the limit (50 µg/kg) recommended by the European Commission (European Union, 2018; European Commission, 2018). Recently, we have published a work related to the determination of BPA in drinking water, BPA was found at the level of 0.29 to 24.88 µg/L in PET bottled water samples whereas in glass bottled water samples the BPA concentration was detected from 4.34 to 41.19 µg/L (AlAmmari et al., 2020). Relatively, the BPA levels were obtained at higher concentration in glass bottled water samples and consistent with the values obtained in the present study.

Consequently, we can propose that the BPA identified in carbonated beverages samples (canned, glass bottled and PET plastic bottled) in the present work is not a threat issue for human health especially in Saud Arabia. Nonetheless, BPA is an endocrine disruptor contaminant which impedes with the eradication and function of usual hormones from human body. BPA can mimic our whole body functions (neuronal growth changes, reproductive system related with reduced fertility, endocrine alterations; cardiovascular and immunological and metabolic illnesses and cancer disease) in a way that could be very risky for human health. These concerns show the necessity of an incessant and precise assessment of all promising origin of BPA, including food products.

4 Conclusion

The aim of the present work was to study the presence of BPA in carbonated beverages of different flavors, origin and packaging materials based on can, glass and PET plastic. The BPA levels were detected at 0.64-11.41 µg/L (canned) 1.92-29.56 µg/L

(glass bottled) and 0.37-21.83 µg/L (PET plastic bottled). So, it can be concluded that BPA amounts in glass bottle beverages are several times greater than those obtained in canned and PET plastic bottled beverage. Nevertheless, levels did not go beyond the particular migration values (0.6 µg/g) and consumption estimations were far-off the acceptable daily intake 50 µg/kg (European Union, 2004, 2018; European Commission, 2018).

The unexpected occurrence of BPA especially in glass bottled beverage at high levels emphasizes the ubiquity of such compound beside the food fabrication chain, far off to the food packaging materials. Therefore, a research is needed to resolve the sources of this endocrine disruptor in beverage samples, in addition to threat assessment studies, are essential to defend human health. However, dietary exploration is a more suitable way to find the major sources of BPA exposure. In future evaluation, will be based on food products and would be more effective to comprise food products packed in various type of packaging materials. This is the first report relating to the occurrence of BPA in canned and, glass and PET bottled beverages in the Saudi Arabian market.

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