

Evaluation of Bis (2-ethylhexyl) phthalate (DEHP) in the PET Bottled Mineral Water of Different Brands and Impact of Heat by GC-MS/MS

Laxmi Kant Bhardwaj (✉ bhardwaj.laxmikant@gmail.com)

AIETSM <https://orcid.org/0000-0001-7518-4199>

Archana Sharma

Amity University

Research Article

Keywords: Bis (2-ethylhexyl) phthalate (DEHP), PET (polyethylene terephthalate), Mineral water, Gas chromatography-mass spectrometry (GC-MS/MS)

DOI: <https://doi.org/10.21203/rs.3.rs-298354/v1>

License: © ⓘ This work is licensed under a Creative Commons Attribution 4.0 International License. [Read Full License](#)

Abstract

Phthalates are one of the ubiquitous contaminants in the environment due to the extensive use in the last few years. They are easily released because they are not chemically bonded to polymers. They migrate into the food during food packing while in water, they migrate during water filling or storage and bottle manufacturing. They are toxic to human health and known as carcinogen/ endocrine disruptors. A total of sixty PET (polyethylene terephthalate) bottled mineral water samples of different six brands were purchased from the local market of Noida, India. These bottles were of two different batch numbers of each brand. Two bottles of each brand with a different batch number were analyzed immediately after purchase while the other eight bottles were analyzed after two and six months when they were stored in sunlight (~ 45 o C) and - 20 o C. The aim of the present study was to determine the migration of DEHP and its impact on storage conditions of PET bottled mineral water in retail stores or homes. We used a gas chromatography-mass spectrometry (GC-MS/MS) for the estimation of DEHP in these samples. We observed that the migration of DEHP was dependent on high temperature and storage time. DEHP was present only in those samples, which were stored in sunlight for two & six months and at - 20 o C for six months. While found below the detection limit in those samples which were analyzed immediately after purchase and stored at - 20 o C for two months.

Introduction

Phthalates (an ester of ortho-phthalic acid) are a group of aromatic chemicals that contain a phenyl ring with two attached acetate groups. They are colorless, odorless liquid which does not evaporate easily. They are manufactured chemicals used to make plastics soft and more flexible. They can be removed easily from plastic resources because the covalent bond is weak between the phthalate and their parent things (Serô dio and Nogueira 2006; Heudorf et al. 2007; Liu et al. 2008; Kim et al. 2011; Serrano et al. 2014). According to Singh and Li (2011) phthalates are plasticizers and softening additives in the manufacture of plastic goods.

Bis (2-ethylhexyl) phthalate is known as diethyl hexyl phthalate (DEHP) and it is the main compound of PET bottled materials which migrates from the PET bottle to the mineral water (Clara et al. 2010). It is the most important congener and is about 50 % of the world's production of phthalates (Cadogan 2002). It can enter into the environment through releases from factories that make or use it. It can also enter into the environment through air, water and household items containing it. A large amount of plastic that contains DEHP is buried in landfill sites. It has been found in groundwater near waste disposal facilities. It usually attaches strongly to the soil while dissolves very slowly into underground water or surface water. The level of DEHP in indoor air could be higher than the outdoor air when coatings and flooring installed recently in a room.

Phthalates are present in plastic products such as toothbrushes, wall coverings, furniture upholstery, toys, paints, adhesives, inks, packaging film and sheets, sheathing for wire and cable, medical tubing and blood storage bags, etc. (ATSDR 2011; Kotowska et al. 2006). The usages of phthalates in plastic products, cosmetics and personal care products make the presence of phthalate in water, wastewater and soil (Horn et al. 2004; Wooten and Smith 2013). They migrate in a low amount into the food, drinking water and beverages from the packaging or manufacturing processes or bottling materials and this is the most probable route of exposure of phthalate. They can enter into the body through food, drinking water, air, skin contact and through medicines. Blood transfusion and kidney dialysis are the other routes of phthalate exposure. Phthalate effluent from the wastewater treatment plant is the main source of phthalate in the aquatic environment (Gao et al. 2014; Berge et al. 2014).

Phthalate may cause developmental toxicity and reproductive effects in animals. They have carcinogenic and teratogenic effects (Becker et al. 2004; Herrero et al. 2015). They exhibit low toxicity with short-term exposure, while exhibit high toxicity with long-term exposure. The USEPA (United States Environmental Agency) and EU (European Union) have banned phthalate in toys and children articles (Biedermann-Brem et al. 2008). Several researchers studied the toxic effect of phthalate on human health and reported that endocrine, liver, cardiovascular, urologic and genital diseases are occurring due to the phthalate (Gomes-Hens and Aguilar-Caballos 2003; Swan et al. 2005; Singh and Li 2011). They are known as hormonally active agents (HAAs) or endocrine-disrupting chemicals (EDCs) because they affect the hormonal system or the endocrine system of the body (U.S. National Toxicology Program 2007). Over the past few decades, phthalate is responsible for the endocrine-mediated adverse effects which was observed in human and wildlife (Amiridou and Voutsas 2011).

Due to the size, strength, ease of transport and low cost of production the most widespread water bottling material is PET (Bach et al. 2013). Bolgar et al. (2015) stated that the plastic material used to manufacture bottles consists of some additives such as catalyzers, accelerators, stabilizers, coupling agents and antioxidants. In the last few decades, the consumption of PET bottled mineral water has increased and has effectively replaced tap water in several developed countries (Petrelli et al. 2006; IBWA 2009; Andra et al. 2011). PET bottled water has emerged as a drinking water, which preserves the original purity of natural mineral water. In many areas where there is a lack of portable public water, PET bottled water is the only water available for human consumption (Guart et al. 2014).

The packaging of the bottle acts as a gas barrier to avoid interaction with the adjacent environment (EU 2011). However, the presence of oxygen at higher temperature promotes thermo-oxidative and thermo-mechanical reactions which enhance the migration of phthalate from PET bottled to the water (Zhang and Ward 1995; Paci and Mantia 1998; Romao et al. 2009). In the presence of oxygen, DEHP can be broken down into carbon dioxide and other simple chemicals in water and soil with the help of microorganisms (ATSDR 2002). The concentration of DEHP in PET bottled mineral water may vary with pH (Montuori et al. 2008), storage time (Biscardi et al. 2003; Criado et al. 2005), storage temperature (Casajuana and Lacorte 2003; Schmid et al. 2008) and exposure to sunlight (Leivadara et al. 2008).

The purpose of this study was to estimate the DEHP migration in the PET bottled mineral water which was stored at different temperature for a different time. We used the GC-MS/MS for the estimation of DEHP. To the best of our information, this study is the first study to estimate the migration of DEHP under normal condition, freezing condition (- 20 °C) and sunlight (~ 45 °C) in the period of July 2018- December 2018.

Materials And Methods

2.1. Sampling: We purchased a total of sixty PET bottled mineral water samples of different six brands (Aquafina, Bisleri, Catch, Divyajal, Kinley and Railneer) from the local market of Noida, India. We purchased ten water bottles of each brand with two different batch numbers. All PET bottled water samples were contained in their original sealed containers. The sampling details are shown in table 1. The first two water bottles of each brand with a different batch number were analyzed immediately after purchase. The other four water bottles of each brand with a different batch number were stored in the refrigerator (at - 20 °C) for 2 months and 6 months, respectively. While another four water bottles of each brand with a different batch number were stored in sunlight (at ~ 45 °C) for the same time.

2.2. Equipment and Reagents: GC-MS/MS (Model No.-7000C, Agilent), HP-5MS column (30 m length×0.25 mm width×0.25 µm film thickness), rotary evaporator (Model No.- R 300, Buchi) and electronic weighing machine (0.01 mg to 220 g, Sartorius) were used for this study.

DEHP standard was purchased from Sigma-Aldrich (Lot No.-BCBR8079V). Dichloromethane and hexane both were procured from Merck. Sodium sulphate and silica gel of mesh size (100/200) were purchased from Fisher Scientific. Prior to use, sodium sulfate was baked at 400 °C for 4 hours and silica gel was triggered at 130 °C for 16 hours. Double distilled water was used throughout the study.

2.3. Standard and Stock Solution Preparation: A stock solution (1000 µg/l) of DEHP was prepared in hexane. Then further standard solutions of different concentrations (0.1 to 50.0 µg/l) were prepared from the stock solution in the same solvent.

• Sample Preparation

2.4.1. Extraction: DEHP was extracted from the PET bottled mineral water samples by using the method USEPA-3510B with slight modification. The bottled water sample was homogenized manually and then half liter of the sample was taken into a glass separatory funnel of one liter. 60 ml of dichloromethane was added into the separatory funnel and then the sample was shaken for 1-2 minutes. After releasing the pressure, the funnel was kept for 10 minutes to separate the organic layer from the aqueous phase. The organic layer of dichloromethane was filtered through the Whatman No.-41 filter paper in 250 ml of the RB (round bottom) flask. This procedure was done two more times and the solvent extract was collected in the same flask. Then the collected solvent was evaporated up to 2 ml at 45 °C by using the rotary evaporator and the final residue was ready for the cleanup process.

2.4.2. Cleanup: The final residue solution was cleaned by using the method USEPA-3630C. A chromatographic glass column (250 mm length × 10 mm width) was prepared with some portion of silica gel (5 g) & sodium sulfate (2 g). The column was preconditioned with the solvent mixture (hexane: dichloromethane, 1:1, v/v). The final residue solution (2 ml) was then transferred onto the chromatographic column. Hexane (50 ml) was added onto the glass column and then was elute out into an RB flask of 250 ml. This process was repeated three more times and the eluted solvent was collected in the same flask. All the eluted solvent was concentrated at 45 °C by using a rotary evaporator and then was re-dissolved in 1 ml of hexane. The sample was ready for the estimation of DEHP by GC-MS/MS.

2.5. Instrumental analysis: The final sample solution (1 µl) was injected into the GC-MS/MS. The separation was done with the help of the capillary column (HP-5MS). Helium was used as a carrier gas with a constant flow of 1 ml/min. The injector temperature was set at 280 °C and the pressure was 10.0 psi. The GC oven temperature was 160 °C, then increased at a rate of 20 °C/min up to 250 °C, again increased at a rate of 1.6 °C/min up to 270 °C which was maintained for 5 min. The total run time was 22.0 min. The detector temperature was set at 300 °C and the makeup gas was nitrogen. The ion source and transfer line temperature were set at 280 °C and 320 °C, respectively.

2.6. Quantitative analysis: Each PET bottled water sample was examined three times and then the average value was taken. One blank sample with each set of water samples was processed. The calibration of the instrument was performed with a DEHP standard range from 0.1 to 50.0 µg/l and the linear correlation (R^2) was > 0.99. The recovery of DEHP was found between 40-100 %, which achieved the recovery limit of method USEPA-1699 (2007). The LOD of the instrument was found 0.05 µg/l.

Results

Some physio-chemical parameters such as pH, TDS (total dissolved solids), conductivity, chloride and nitrate (NO_3) were measured with the estimation of phthalate (DEHP) in the PET bottled mineral water samples. These samples were agreeable in odour and taste in all the storage conditions. The result is shown in tables 2, 3, 4, 5 & 6. The graphical representation of the changing of concentrations is shown in figures 1, 2, 3, 4, 5 & 6.

3.1. Analysis after purchase: The pH was in the range of 6.74-6.98. The TDS was varied from 4.90-168.33 mg/l and the range of conductivity was found from 5.60-278.00 µS/cm. The chloride was in the range of 4.5-15.50 mg/l and nitrate was in the range of 2.78-27.12 mg/l. DEHP was found below the detection limit in all the samples.

3.2. Analysis after two months: The pH was in the range of 6.51-6.78 at ~ 45 °C and 6.73-6.96 at - 20 °C. The TDS was from 6.16-170.21 mg/l at ~ 45 °C and 4.88-168.10 mg/l at - 20 °C. The conductivity was in the range of 6.20-280.56 µS/cm at ~ 45 °C and 5.62-277.87 µS/cm at - 20 °C. The chloride was in the range of 4.00-15.00 mg/l at ~ 45 °C and 4.50-15.50 mg/l at - 20 °C. Nitrate was varied from 2.98-28.86 mg/l at ~ 45 °C and 2.77-27.00 mg/l at - 20 °C. DEHP was found from 0.52-3.26 µg/l at ~ 45 °C and below the detection limit at - 20 °C.

3.3. Analysis after six months: The pH was varied from 6.12-6.36 at ~ 45 °C and 6.57-6.81 at - 20 °C. The TDS was in the range of 8.28-180.45 mg/l at ~ 45 °C and 5.98-174.56 mg/l at - 20 °C. The conductivity was from 8.11-286.32 µS/cm at ~ 45 °C and 6.82-280.54 µS/cm at - 20 °C. The chloride was from 2.50-

13.50 mg/l at ~ 45 °C and 4.00-15.00 mg/l at - 20 °C. Nitrate was in the range of 3.76-35.22 mg/l at ~ 45 °C and 3.12-28.10 mg/l at - 20 °C. DEHP was found from 1.10-5.39 µg/l at ~ 45 °C and 0.31-2.66 µg/l at - 20 °C.

Discussion

The result indicates that the value of EC, TDS and NO₃ is increasing in the PET bottled mineral water samples with the increase of temperature. While the value of pH and Cl⁻ is decreasing with the increase of temperature. The increase of TDS and EC may be due to the leaching of metals (inorganic compounds) and ions (organic compounds) from the PET bottle to the water. Hansen et al. (2006) and Fiket et al. (2007) studied the leaching of metals from PET bottled to water and stated that it was the main reason to increase the TDS & EC in water. The decrease of chloride ions may be due to the chlorination reaction (Muhamad et al. 2011) and the decrease of pH is due to the oxidation of organic compounds through the photodegradation by sunlight (Monarca et al. 1994). Temperature influences on the leaching of organic and inorganic compounds from PET bottled to water (Villain et al. 1995; Pinto and Reali 2009). We found the highest TDS, conductivity and chloride in the Catch brand followed by Railneer, Bisleri, Divyajal, Kinley & Aquafina. The highest nitrate was found in the Railneer brand followed by Divyajal, Bisleri, Catch, Kinley and Aquafina.

Jeddi et al. (2015) analyzed PET bottled water samples immediately after purchase and stated that there was no significant correlation between the DEHP concentration and the physicochemical properties of the water samples. While Dumitrascu (2012) reported that the migration of DEHP from PET bottled to water depends on the pH. DEHP was the most abundant phthalate in the PET bottled water samples and acts as a carcinogenic agent (Schmid et al. 2008; Greifenstein et al. 2013; ; Keresztes et al. 2013). Although due to analytical difficulties in achieving a low detection limit, there is a lack of data on the leaching of DEHP in PET bottled mineral water. Moreover, some studies have done and reported the DEHP concentration from 0.35 µg/l to 8.8 µg/l in PET bottled water samples which were kept at different temperature for the different time (Psillakis and Kalogerakis 2003; Bosnir et al. 2007; Amiridou and Voutsas 2011; Al-Saleh et al. 2011).

Casajuana and Lacorte (2003) had reported the highest concentration of DEHP (<0.002-0.188 µg/l) in PET bottled water when the samples were stored in outdoors for 10 weeks at 30 °C. Schmid et al. (2008) and Muhamad et al. (2011) had reported the DEHP concentration (0.10-0.71 µg/l) in PET bottled water samples after 17 hr of incubation in darkness/sunlight at room temperature and at 60 °C. Leivadara et al. (2008) had reported the DEHP concentration (<0.002-6.8 µg/l) in bottled water samples when the samples were stored at 30 °C for 3 months. Keresztes et al. (2013) had studied the leaching of DEHP from PET bottles to water and detected the DEHP concentration up to 1.7 µg/l. Guart et al. (2014) had reported the phthalate concentration from 0.022 to 20.5 µg/l in 12 fresh PET bottled water samples and 0.635 to 13.0 µg/l in 37 one year stored samples. Jeddi et al. (2015) reported the lowest (0.32 µg/l) and highest (0.92 µg/l) concentration of DEHP at -18 °C and 40 °C respectively.

Wegelin et al. (2001) reported the terephthalate monomers and dimers, which were the PET degradation products in the presence of sunlight. Monarca et al. (1994) stated that phthalate esters are produced from the organic compounds after oxidation through photodegradation in sunlight. Peterson (2003) has described that photolysis is the significant pathway for abiotic degradation of phthalates in water. The exposure to phthalates causes developmental and reproductive toxicities in rodents (Martino-Andrade and Chahoud 2010; Meeker 2009). Plastic components migrate in low quantities into the fresh water during the bottle manufacturing or bottling process and depend on the water brand. The bottle manufacturing process could also be a source of plasticizers when the polymerization process has not been completed.

After production, the presence of DEHP in PET bottled water may be due to the contamination in the bottling plant or contamination in water treatment facilities and the source of water. The effect of environmental factors such as high temperature, etc. on the PET bottled water samples from manufacture to purchase cannot be excluded (Al-Saleh et al. 2011; Amiridou and Voutsas 2011; Keresztes et al. 2013). Lin et al. (2000) stated that PET degradation is temperature-dependent and it occurs more rapidly at high temperature. In the present study, We have reported the highest migration of DEHP from PET bottled to water samples when the samples were stored at sunlight (~ 45 °C) for six months and two months. While the migration of DEHP was observed below detection limits when samples were analyzed immediately after purchase and stored at - 20 °C for two months. This study confirmed that the migration of DEHP is higher at high temperature while lower at the low temperature. The highest migration of DEHP was observed in the Railneer brand (2.97-5.39 µg/l) followed by Catch (2.10-4.90 µg/l), Bisleri (1.67-3.87 µg/l), Divyajal (0.95-2.65 µg/l), Kinley (0.76-1.73 µg/l) and Aquafina (0.52-1.31 µg/l) at ~ 45 °C after two & six months. While the DEHP concentration was found below the detection limit when the samples were analyzed after purchase and after two months (stored at - 20 °C). Some amount of DEHP was observed in the Railneer brand (2.03-2.66 µg/l) followed by Catch (1.87-1.99 µg/l), Bisleri (1.03-1.25 µg/l), Divyajal (0.81-0.89 µg/l), Kinley (0.51-0.55 µg/l) and Aquafina (0.31-0.38 µg/l) after six months when the samples were stored at - 20 °C.

As per the USEPA and WHO a maximum permissible limit of DEHP in drinking water is 6-8 µg/l (WHO 2008; USEPA 2009). Wormuth et al. (2006) and Jeddi et al. (2015) stated that phthalate exposure through water is reduced with increasing age because water intake is higher in children as compared to adults. As per the IARC (The International Agency for Research on Cancer), only DEHP is carcinogenic to humans in all the phthalate esters (USEPA 2012c). The cancer risk due to DEHP exposure via water intake stored at 40 °C was greater than for the other storage conditions while the least carcinogenic risk from DEHP was observed in the bottled water stored under freezing conditions (-18 °C) (Jeddi et al. 2015). Due to the DEHP concentration, the carcinogenic risk was tremendously below the accepted risk level because the DEHP in water corresponding to cancer risk in 1,000,000 was 3 µg/l (USEPA 2012c). Finally, the carcinogenic risk posed by the highest concentration of DEHP in PET bottled water samples is negligible. Bolgar et al. (2015) reported that DEHA can be considered an effective replacement for DEHP.

Conclusions

PET material is used worldwide as a container for bottled water; even it is used in which area where the drinking water problem occurs. In the present study, PET bottled mineral water samples were studied in different storage conditions which give an answer to public questions about the safety of bottled water. The different storage conditions such as temperature and time are the main factors which affect the migration of DEHP from PET bottles to water. DEHP was estimated in the PET bottled water samples by GC-MS/MS.

After comparing the results of DEHP in PET bottled water before and after storage, We conclude that poor storage conditions cause an increase in the concentration of DEHP. The migration of DEHP from the PET bottle to water depended on the pH of the water samples and increases with the decreasing pH. DEHP migration was increased with the increasing temperature and time while migration was observed low when the samples were stored at low temperatures (>25 °C). The migration of DEHP may depend on the brand because water characteristics and bottling process vary from brand to brand. We observed that DEHP was detected in six-months & two months stored water samples which were stored in sunlight (~ 45 °C). Some amount of DEHP was also observed in samples which were stored for six-months at - 20 °C.

In spite of the consumption of PET bottled water was more in developing and developed countries the detected concentration of DEHP was far below the toxic levels. Therefore, the main conclusion of this study is that all common conditions of PET bottled water stored in retail supermarkets, outlets and homes are nontoxic/safe for consumers. Further analysis of phthalates is required for the prevention of the health effects of chemical exposure considering the development of PET bottled water consumption.

Declarations

6.1. Ethics approval and consent to participate: Not applicable.

6.2. Consent for publication: Not applicable.

6.3. Availability of data and materials: Not applicable.

6.4. Competing interest: The authors declare that they have no competing interests.

6.5. Funding: This study was not supported by any funding agency.

6.6. Author's contributions: LKB analyzed all PET bottled water samples for the different parameters. AS was a major contributor in writing the manuscript. All authors read and approved the final manuscript.

References

ATSDR -Agency for toxic substances and disease registry (2002) Toxicological Profile for Di (2-ethylhexyl) phthalate (DEHP). Public Health Service, U.S. Department of Health and Human Services, Atlanta, GA.

ATSDR (2011) <http://www.atsdr.cdc.gov> accessed on December, 16.

Al-Saleh I, Shinwari N, Alsabbaheen A (2011) Phthalates residues in plastic bottled waters. *The Journal of toxicological sciences* 36(4): 469-478.

Amiridou D, Voutsas D (2011) Alkylphenols and phthalates in bottled waters. *Journal of hazardous materials* 185(1): 281-286.

Andra SS, Makris KC, Shine JP (2011) Frequency of use controls chemical leaching from drinking-water containers subject to disinfection. *Water research* 45(20): 6677-6687.

Bach C, Dauchy X, Severin I, Munoz JF, Etienne S, Chagnon MC (2013) Effect of temperature on the release of intentionally and non-intentionally added substances from polyethylene terephthalate (PET) bottles into water: Chemical analysis and potential toxicity. *Food Chemistry* 139(1-4): 672-680.

Becker K, Seiwert M, Angerer J, Heger W, Koch HM, Nagorka R, Roßkamp E, Schlüter C, Seifert B, Ullrich D (2004) DEHP metabolites in urine of children and DEHP in house dust. *International journal of hygiene and environmental health* 207(5): 409-417.

Bergé A, Gasperi J, Rocher V, Gras L, Coursimault A, Moilleron R (2014) Phthalates and alkylphenols in industrial and domestic effluents: Case of Paris conurbation (France). *Science of the total environment* 488: 26-35.

Biedermann-Brem S, Biedermann M, Pfenninger S, Bauer M, Altkofer W, Rieger K, Hauri U, Droz C, Grob K (2008) Plasticizers in PVC toys and childcare products: What succeeds the phthalates? Market survey 2007. *Chromatographia* 68(3-4): 227-234.

Biscardi D, Monarca S, De Fusco R, Senatore F, Poli P, Buschini A, Rossi C, Zani C (2003) Evaluation of the migration of mutagens/carcinogens from PET bottles into mineral water by Tradescantia/micronuclei test, Comet assay on leukocytes and GC/MS. *Science of the Total Environment* 302(1-3): 101-108.

Bolgar M, Hubball J, Groeger J, Meronek S (2015) Handbook for the chemical analysis of plastic and polymer additives. CRC Press.

Bošnić J, Puntarić D, Galić A, Škes I, Dijanić T, Klarić M, Grgić M, Čurković M, Šmit Z (2007) Migration of phthalates from plastic containers into soft drinks and mineral water. *Food Technology and Biotechnology* 45(1): 91-95.

Cadogan D (2002) Health and environmental impact of phthalates. *Plastics, Additives and Compounding* 4(6): 28-29.

- Casajuana N, Lacorte S (2003) Presence and release of phthalic esters and other endocrine disrupting compounds in drinking water. *Chromatographia* 57(9-10): 649-655.
- Clara M, Windhofer G, Hartl W, Braun K, Simon M, Gans O, Scheffknecht C, Chovanec A (2010) Occurrence of phthalates in surface runoff, untreated and treated wastewater and fate during wastewater treatment. *Chemosphere* 78(9): 1078-1084.
- Criado MV, Pinto VEF, Badessari A, Cabral D (2005) Conditions that regulate the growth of moulds inoculated into bottled mineral water. *International Journal of Food Microbiology* 99(3): 343-349.
- Dumitrascu I (2012) Determination of phthalates from bottled water by GC-MS. *Aerul si Apa. Componente ale Mediului* 337.
- EU (2011) Commission Regulation (EU) No 10/2011 of 14 January 2011 on plastic materials and articles intended to come in contact with food. Official journal of the European Commission.
- Fiket Ž, Roje V, Mikac N, Kniewald G (2007) Determination of arsenic and other trace elements in bottled waters by high resolution inductively coupled plasma mass spectrometry. *Croatica chemica acta* 80(1): 91-100.
- Gao D, Li Z, Wen Z, Ren N (2014) Occurrence and fate of phthalate esters in full-scale domestic wastewater treatment plants and their impact on receiving waters along the Songhua River in China. *Chemosphere* 95: 24-32.
- Gomez-Hens A, Aguilar-Caballeros MP (2003) Social and economic interest in the control of phthalic acid esters. *TrAC Trends in Analytical Chemistry* 22(11): 847-857.
- Greifenstein M, White DW, Stubner A, Hout J, Whelton AJ (2013) Impact of temperature and storage duration on the chemical and odor quality of military packaged water in polyethylene terephthalate bottles. *Science of the total environment* 456: 376-383.
- Guart A, Bono-Blay F, Borrell A, Lacorte S (2014) Effect of bottling and storage on the migration of plastic constituents in Spanish bottled waters. *Food chemistry* 156: 73-80.
- Hansen HR, Pergantis SA (2006) Detection of antimony species in citrus juices and drinking water stored in PET containers. *Journal of Analytical Atomic Spectrometry* 21(8): 731-733.
- Herrero-Latorre C, Barciela-García J, García-Martín S, Peña-Crecente RM, Otárola-Jiménez J (2015) Magnetic solid-phase extraction using carbon nanotubes as sorbents: a review. *Analytica Chimica Acta* 892: 10-26.
- Heudorf U, Mersch-Sundermann V, Angerer J (2007) Phthalates: toxicology and exposure. *International journal of hygiene and environmental health* 210(5): 623-634.
- Horn O, Nalli S, Cooper D, Nicell J (2004) Plasticizer metabolites in the environment. *Water Research* 38(17): 3693-3698.
- IBWA (2009) [International bottled water association. Bottled water vs Tap water.](#)
- Jeddi MZ, Rastkari N, Ahmadvaniha R, Yunesian M (2015) Concentrations of phthalates in bottled water under common storage conditions: Do they pose a health risk to children?. *Food research international* 69: 256-265.
- Keresztes S, Tatár E, Czégény Z, Záray G, Mihucz VG (2013) Study on the leaching of phthalates from polyethylene terephthalate bottles into mineral water. *Science of the total environment* 458: 451-458.
- Kim HH, Yang JY, Kim SD, Yang SH, Lee CS, Shin DC, Lim YW (2011) Health risks assessment in children for phthalate exposure associated with childcare facilities and indoor playgrounds. *Environmental health and toxicology* 26.
- Kotowska U, Garbowska K, Isidorov VA (2006) Distribution coefficients of phthalates between absorption fiber and water and its using in quantitative analysis. *Analytica chimica acta* 560(1-2): 110-117.
- Leivadara SV, Nikolaou AD, Lekkas TD (2008) Determination of organic compounds in bottled waters. *Food Chemistry* 108(1): 277-286.
- Lin JF, Ho CF, Huang SK (2000) Studies on curing kinetics and total thermal degradation of the modified epoxy copolymer with penta-coordinated phosphate as a tribranched junction. *Journal of applied polymer science* 77(4): 719-732.
- Liu HC, Den W, Chan SF, Kin KT (2008) Analysis of trace contamination of phthalate esters in ultrapure water using a modified solid-phase extraction procedure and automated thermal desorption-gas chromatography/mass spectrometry. *Journal of Chromatography A* 1188(2): 286-294.
- Martino-Andrade AJ, Chahoud I (2010) Reproductive toxicity of phthalate esters. *Molecular nutrition & food research* 54(1): 148-157.
- Meeker JD, Sathyanarayana S, Swan SH (2009) Phthalates and other additives in plastics: human exposure and associated health outcomes. *Philosophical Transactions of the Royal Society B: Biological Sciences* 364(1526): 2097-2113.

- Monarca S, Fusco RD, Biscardi D, Feo VD, Pasquini R, Fatigoni C, Moretti M, Zanardini A (1994) Studies of migration of potentially genotoxic compounds into water stored in PET bottles. *Food and chemical toxicology* 32(9): 783-788.
- Montuori P, Jover E, Morgantini M, Bayona JM, Triassi M (2008) Assessing human exposure to phthalic acid and phthalate esters from mineral water stored in polyethylene terephthalate and glass bottles. *Food Additives and Contaminants* 25(4): 511-518.
- Muhamad SG, Esmail LS, Hasan SH (2011) Effect of storage temperature and sunlight exposure on the physicochemical properties of bottled water in Kurdistan region-Iraq. *Journal of Applied Sciences and Environmental Management* 15(1).
- Paci M, La Mantia FP (1998) Competition between degradation and chain extension during processing of reclaimed poly (ethylene terephthalate). *Polymer degradation and Stability* 61(3): 417-420.
- Petrelli F, Grappasonni I, Morichetti L, Cocchioni M (2006) Evoluzione normativa Della disciplina delle acque destinate al consumo umano. *Annali di Igiene* 15: 923-931.
- Peterson DR, Staples CA (2003) Degradation of phthalate esters in the environment. In *Series Anthropogenic Compounds* 85-124.
- Pinto B, Reali D (2009) Screening of estrogen-like activity of mineral water stored in PET bottles. *International Journal of Hygiene and Environmental Health* 212(2): 228-232.
- Psillakis E, Kalogerakis N (2003) Hollow-fibre liquid-phase microextraction of phthalate esters from water. *Journal of Chromatography A* 999(1-2): 145-153.
- Romao W, Franco MF, Corilo YE, Eberlin MN, Spinace MA, De-Paoli MA (2009) Poly (ethylene terephthalate) thermo-mechanical and thermo-oxidative degradation mechanisms. *Polymer Degradation and Stability* 94(10): 1849-1859.
- Schmid P, Kohler M, Meierhofer R, Luzi S, Wegelin M (2008) Does the reuse of PET bottles during solar water disinfection pose a health risk due to the migration of plasticisers and other chemicals into the water?. *Water research* 42(20): 5054-5060.
- Serôdio P, Nogueira JMF (2006) Considerations on ultra-trace analysis of phthalates in drinking water. *Water research* 40(13): 2572-2582.
- Serrano SE, Braun J, Trasande L, Dills R, Sathyanarayana S (2014) Phthalates and diet: a review of the food monitoring and epidemiology data. *Environmental Health* 13(1): 43.
- Singh S, Li SSL (2011) Phthalates: toxicogenomics and inferred human diseases. *Genomics* 97(3): 148-157.
- Swan SH, Main KM, Liu F, Stewart SL, Kruse RL, Calafat AM, Mao CS, Redmon JB, Ternand CL, Sullivan S, Teague JL (2005) Decrease in anogenital distance among male infants with prenatal phthalate exposure. *Environmental health perspectives* 113(8): 1056-1061.
- US National Toxicology Program (2007) Center for the evaluation of risks to human reproduction. "NTP-CERHR reports and monographs." <http://cerhr.niehs.nih.gov/reports/index.html>.
- USEPA (U.S. Environmental Protection Agency) (2012) Di(2-ethylhexyl) phthalate (DEHP) (CASRN 117-81-7). Available <http://www.epa.gov/iris/subst/0014.htm> ([accessed 15 March 2012])
- USEPA (2009) Drinking water contaminants. U.S. Environmental Protection Agency, EPA (816-F-09-0004, May 2009).
- USEPA (2007) Method 1699: Pesticides in Water, Soil, Sediment, Biosolids and Tissue by HRGC/HRMS.
- USEPA (1994) Method 3510B: Separatory Funnel Liquid Liquid Extraction. Revision 2, September 1994.
- USEPA (1996) Method 3630C: Silica Gel Cleanup. Revision 3, December 1996.
- Villain F, Coudane J, Vert M (1995) Thermal degradation of polyethylene terephthalate: study of polymer stabilization. *Polymer degradation and stability* 49(3): 393-397.
- Wegelin M, Canonica S, Alder C, Marazuela D, Suter MF, Bucheli TD, Haefliger OP, Zenobi R, McGuigan KG, Kelly MT, Ibrahim P (2001) Does sunlight change the material and content of polyethylene terephthalate (PET) bottles?. *Journal of Water Supply: Research and Technology—AQUA* 50(3): 125-135.
- Wooten KJ, Smith PN (2013) Canine toys and training devices as sources of exposure to phthalates and bisphenol A: quantitation of chemicals in leachate and in vitro screening for endocrine activity. *Chemosphere* 93(10): 2245-2253.
- World Health Organization (2008) Guidelines for Drinking-water Quality 3rd Ed. Incorporating the First and Second Addenda Volume 1, Recommendations. World Health Organization, Geneva, Switzerland. Available from: www.who.int/water_sanitation_health/dwq/gdwq3rev/en/index.html.
- Wormuth M, Scheringer M, Vollenweider M, Hungerbühler K (2006) What are the sources of exposure to eight frequently used phthalic acid esters in Europeans?. *Risk Analysis* 26(3): 803-824.
- Zhang H, Ward IM (1995) Kinetics of hydrolytic degradation of poly (ethylene naphthalene-2, 6-dicarboxylate). *Macromolecules* 28(23): 7622-7629.

Tables

Table 1: Identification of Samples with Batch No. and Year of Packaging

S. No.	Brand Name	Batch No.	Packing Year
1	Aquafina	7476D12J18	2018
		7476D18J18	
2	Bisleri	297	
		341	
3	Catch	100W008	
		100W154	
4	Divyajal	UP0330133	
		UP0330134	
5	Kinley	H30E8E11	
		H30E8E12	
6	Railneer	2310	
		2311	

Table 2: Characteristics of the Examined PET Bottled Water Samples after Purchase

Parameters	Brands Name with Sample ID											
	Aquafina		Bisleri		Catch		Divyajal		Kinley		Railneer	
	7476D12J18	7476D18J18	297	341	100W008	100W154	UP0330133	UP0330134	H30E8E11	H30E8E12	2310	2311
Appearance	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable
Color	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable
pH	6.94	6.98	6.77	6.74	6.89	6.90	6.91	6.96	6.92	6.90	6.84	6.80
Total Dissolved Solids (mg/l)	5.00	4.90	85.30	88.45	165.78	168.33	68.60	72.40	22.80	20.40	129.60	130.90
Total Hardness (mg/l)	5.60	5.81	178.70	180.40	275.90	278.00	136.76	139.40	41.80	39.80	246.60	243.96
Calcium (mg/l)	4.50	5.00	7.50	8.00	15.00	15.50	7.00	7.50	5.00	4.50	9.50	10.00
Magnesium (mg/l)	2.78	3.02	15.56	16.04	4.65	4.87	18.43	18.68	4.22	4.56	26.87	27.12
Fluoride ($\mu\text{g/l}$)	< BDL	< BDL	< BDL	< BDL	< BDL	< BDL	< BDL	< BDL	< BDL	< BDL	< BDL	< BDL

Table 3: Characteristics of the Examined PET Bottled Water Samples after Two Months at ~ 45 °C

Parameters	Brands Name with Sample ID											
	Aquafina		Bisleri		Catch		Divyajal		Kinley		Railneer	
	7476D12J18	7476D18J18	297	341	100W008	100W154	UP0330133	UP0330134	H30E8E11	H30E8E12	2310	2311
	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable
	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable
	6.72	6.78	6.53	6.51	6.67	6.72	6.72	6.76	6.72	6.69	6.68	6.63
mg/l)	6.56	6.16	87.45	90.33	167.23	170.21	71.30	74.22	24.60	22.10	131.90	133.70
ctivity n)	6.20	6.76	181.76	184.80	278.01	280.56	139.42	141.98	43.30	41.65	248.66	245.21
de	4.00	4.50	7.00	7.50	14.50	15.00	6.50	7.00	4.50	4.00	9.00	9.50
e	2.98	3.16	15.89	16.22	4.73	4.99	19.04	19.78	4.88	4.99	28.12	28.86
(µg/l)	0.58	0.52	1.87	1.67	2.1	2.5	0.95	0.98	0.88	0.76	3.26	2.97

Table 4: Characteristics of the Examined PET Bottled Water Samples after Two Months at - 20 °C

Parameters	Brands Name with Sample ID											
	Aquafina		Bisleri		Catch		Divyajal		Kinley		Railneer	
	7476D12J18	7476D18J18	297	341	100W008	100W154	UP0330133	UP0330134	H30E8E11	H30E8E12	2310	2311
	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable
	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable
	6.94	6.96	6.76	6.73	6.87	6.90	6.90	6.95	6.91	6.90	6.83	6.80
mg/l)	5.00	4.88	85.28	88.21	165.33	168.10	68.40	72.20	22.50	20.20	129.41	130.53
ctivity m)	5.62	5.80	178.58	180.32	275.88	277.87	136.63	139.33	41.77	39.78	246.01	243.78
ide)	4.50	5.00	7.50	8.00	15.00	15.50	7.00	7.50	5.00	4.50	9.50	10.00
e)	2.77	2.98	15.53	15.93	4.61	4.80	18.38	18.59	4.12	4.51	26.65	27.00
(µg/l)	< BDL	< BDL	< BDL	< BDL	< BDL	< BDL	< BDL	< BDL	< BDL	< BDL	< BDL	< BDL

Table 5: Characteristics of the Examined PET Bottled Water Samples after Six Months at ~ 45 °C

S. No.	Parameters	Brands Name with Sample ID											
		Aquaflina		Bisleri		Catch		Divyajal		Kinley		Railneer	
		7476D12J18	7476D18J18	297	341	100W008	100W154	UP0330133	UP0330134	H30E8E11	H30E8E12	2310	2311
1	Odour	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable
2	Taste	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable
3	pH	6.31	6.36	6.12	6.14	6.28	6.31	6.31	6.36	6.35	6.32	6.24	
4	TDS (mg/l)	8.90	8.28	89.65	93.06	178.34	180.45	77.98	80.60	29.50	27.40	138.21	132.54
5	Conductivity (µS/cm)	8.11	8.34	189.32	191.80	283.56	286.32	144.98	147.12	49.02	47.54	253.80	247.66
6	Chloride (mg/l)	2.50	3.00	5.50	6.00	13.00	13.50	5.00	5.50	3.00	2.50	7.50	9.50
7	Nitrate (mg/l)	3.76	3.99	16.53	16.87	5.87	6.03	21.33	21.87	6.00	6.22	34.12	28.10
8	DEHP (µg/l)	1.31	1.10	3.67	3.87	4.76	4.90	2.16	2.65	1.73	1.62	5.25	2.66

Table 6: Characteristics of the Examined PET Bottled Water Samples after Six Months at - 20 °C

Parameters	Brands Name with Sample ID											
	Aquaflina		Bisleri		Catch		Divyajal		Kinley		Railneer	
	7476D12J18	7476D18J18	297	341	100W008	100W154	UP0330133	UP0330134	H30E8E11	H30E8E12	2310	2311
Odour	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable
Taste	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable	Agreeable
pH	6.75	6.81	6.59	6.57	6.69	6.71	6.69	6.74	6.70	6.69	6.66	6.62
TDS (mg/l)	6.50	5.98	88.77	90.44	172.98	174.56	71.45	78.62	24.21	22.25	131.20	132.54
Conductivity (µS/cm)	6.82	7.01	183.50	188.80	278.99	280.54	140.80	144.78	45.64	44.20	249.01	247.66
Chloride (mg/l)	4.00	4.50	7.00	7.50	14.50	15.00	6.50	7.00	4.50	4.00	9.00	9.50
Nitrate (mg/l)	3.12	3.33	15.88	16.03	5.02	5.65	18.88	19.05	4.87	5.00	27.56	28.10
DEHP (µg/l)	0.31	0.38	1.03	1.25	1.87	1.99	0.81	0.89	0.55	0.51	2.03	2.66

Figures

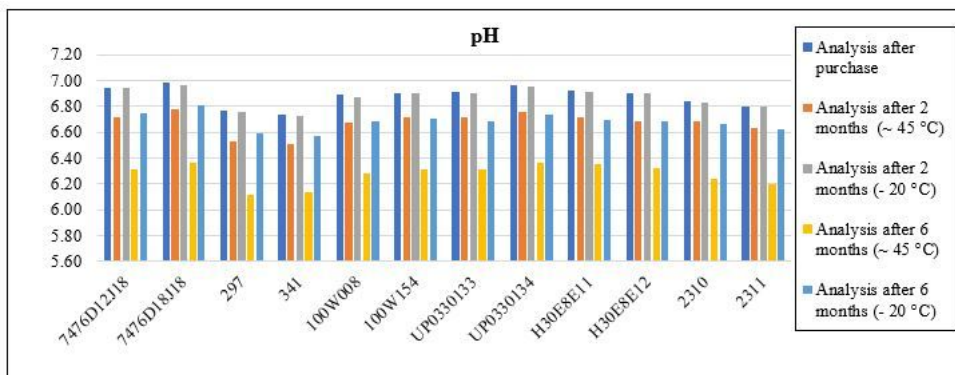


Figure 1

The Effect of Time Duration (Immediately, 2 & 6 Months) at Different Storage Conditions on pH

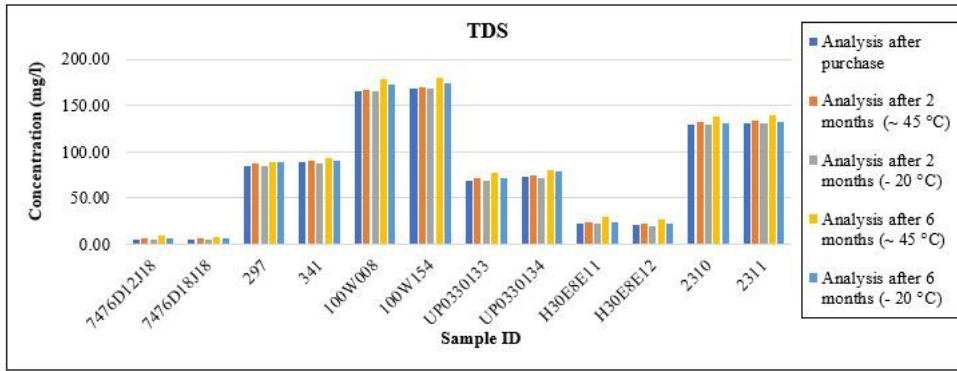


Figure 2

The Effect of Time Duration (Immediately, 2 & 6 Months) at Different Storage Conditions on TDS

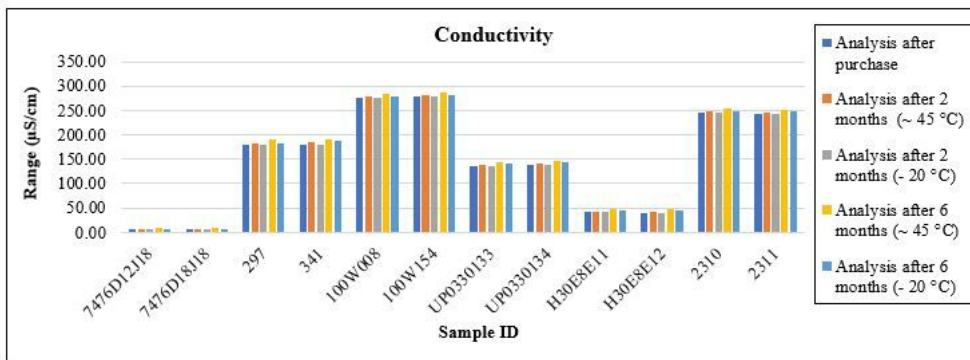


Figure 3

The Effect of Time Duration (Immediately, 2 & 6 Months) at Different Storage Conditions on Conductivity

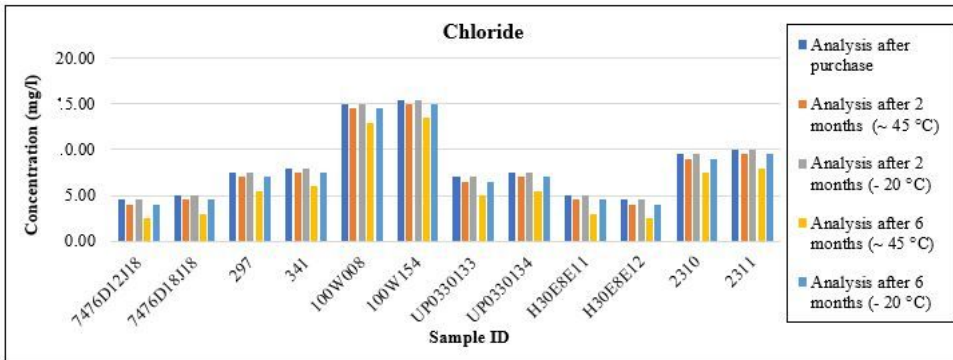


Figure 4

The Effect of Time Duration (Immediately, 2 & 6 Months) at Different Storage Conditions on Conductivity

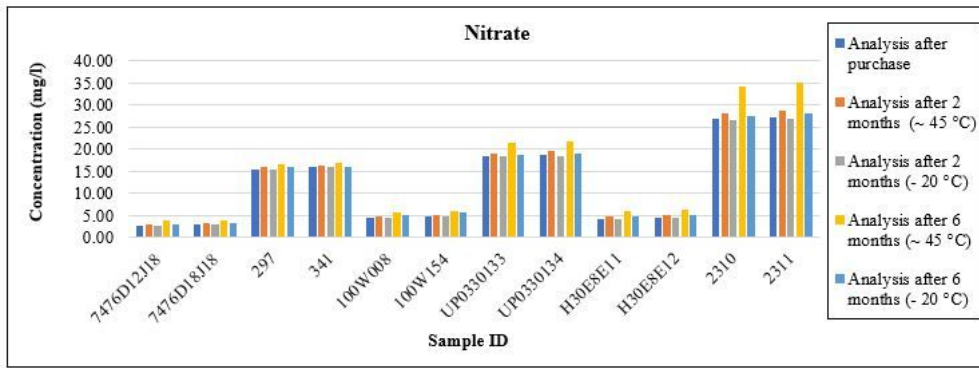


Figure 5

The Effect of Time Duration (Immediately, 2 & 6 Months) at Different Storage Conditions on Nitrate

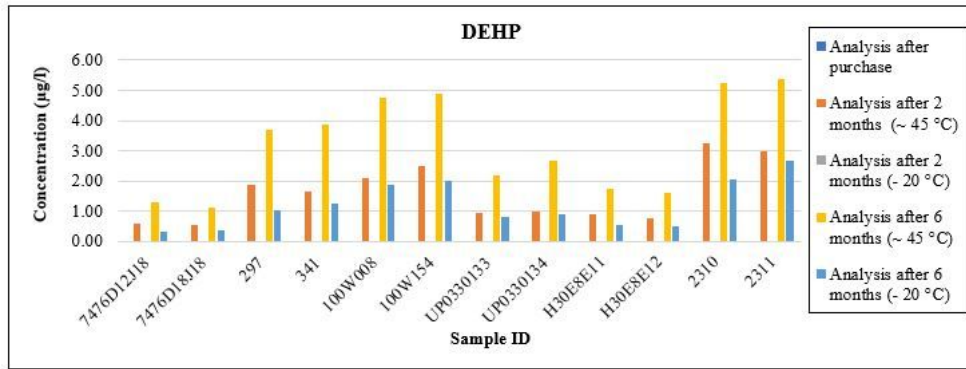


Figure 6

The Effect of Time Duration (Immediately, 2 & 6 Months) at Different Storage Conditions on The Migration of DEHP