

Migration of Plasticizers from Polyethylene Terephthalate Bottles to Vinegar and Water via different Temperature and Storage Periods

Seham A Orabi¹, Taha A Abd-ElRazic², and Hanaa H. El Sayed^{1*}

1-Department of Food Chemistry and Metabolism, National Nutrition Institute (NNI) - Cairo, Egypt.

2-Department of Environmental Basic Sciences, Collage of Environmental Studies and Research, Ain Shams University - Cairo, Egypt.

*Corresponding author: Hanaa H Elsaysd, Email: Hanaa_Hamad2003@yahoo.com Mobil/ +02 01008825869

SUMMARY

Plasticizers (diethyl phthalate (DEP), di-butyl phthalate (DBP), benzyl butyl phthalate (BBP), and diethyl hexyl phthalate (DEHP) are widely used in the manufacture of plastics. Phthalates are not covalently bound to the plastic matrix and can leach into food during packaging. This study investigated the migration of phthalate derivatives from Polyethylene terephthalate (PET) bottles into their water and vinegar under different storage conditions. Gas chromatography (GC) with a mass spectrometry (MS) system is used. The results showed that the highest migration level was DEHP levels after 4 months at 40°C reached 62.5 and 51.9 µg/L in bottled water and vinegar these values were 10.4 and 8.65 times higher than the upper limit for maximum contamination level (MCL) in drinking water as regulated by U.S.EPA, which equal 6.0 µg/L for DEHP.

Keywords: Plasticizers, Migration, Storage, temperatures

FOREWORD

Carvalho et al., (2021) reported that plastics have some advantages for food packaging, there are flexible and inexpensive. The use of plastics in the food industry, such as in packaging, containers, and packaging materials for beverages and processed and raw foods, raises serious issues of food and environmental safety (**Crinnion, 2010**).

Polyethylene terephthalate (PET) is used as a plasticizer and additive to plastics in food industries. The use of PET bottles increased in recent years given their durability, chemical and physical stability, and favorable economic costs. Its use is mainly for food and beverage packaging (**Carneado, 2021**).

Huang et al., (2021) focused on PET as phthalic acid esters (PAEs) are a class of lipophilic chemicals used as plasticizers and additives to improve various products' mechanical extensibility and flexibility. Their characteristic properties, such as their good insulation, high strength, excellent corrosion resistance, low cost, and

ease of fabrication (**He et al., 2019 and Chi et al., 2017**).

The esterification of phthalic anhydride with alcohols of carbon chain lengths is produced phthalates. These reactions run in two successive steps in a closed system. The first step is rapid, irreversible, and results in the formation of monoesters by alcohols of the phthalic acid. The second step involves the formation of the di-ester in the presence of an acid catalyst at temperatures in the range of 140- 165°C or in the presence of atmospheric catalysts at temperatures in the range of 200-250°C. Vacuum distillation or activated charcoal is used to purify phthalate (**Skrzypek et al., 2008**).

Moreover, as heterodoxy is always in progress, it cannot be excluded that other monomers can be used after their complete risk assessment following regulatory requirements. The substance diethyl [[3,5-bis(1,1-dimethyl ethyl)-4-hydroxyphenyl]methyl] phosphonate, considered a comonomer to enhance heat resistance of PET, received recently a positive opinion from the European Food Safety Authority (**EFSA 2016 a**). Phthalates are not covalently bound to the plastic

matrix and can thus leach into food during its production, packaging, or preparation (Tice, 2000).

PAEs have environmental and biomedical pollutants, which may enter the human body through airborne, skin contact, and food chain, constituting possibility health and ecological system threats (Gani, et al., 2017). Epidemiologic studies found that early phthalates exposure could induce significant neuro-developmental damage (Miodovnik, et al., 2014). Some PAEs have been proven to possess reproductive and developmental toxicities to animals and are suspected of causing endocrine-disrupting effects in humans (Wang et al., 2015). Moreover, Phthalates are fat-soluble compounds suspected to interfere with the endocrine system, including androgens, estrogens, and the thyroid-hormone system (Mankidy et al., 2012). Compounds disturbing the endocrine system have been associated with growth during infancy and early childhood and with the development of the reproductive tract (Divall and Opin 2013).

Several agencies of food safety and food standards have imposed restrictions on the use of phthalates in food packaging. The European Union established limits within Regulation No. 10/2011 and modified many compounds used in packaging and fixed specific migration limits for single contaminants or groups of contaminants based on toxicological data (EU) No 10/2011 (EU, 2011).

The present study was carried out to investigate the migration of phthalate derivatives [dimethyl phthalate (DMP), diethyl phthalate (DEP), di-butyl phthalate (DBP), benzyl butyl phthalate (BBP), diethyl hexyl phthalate (DEHP), and di-n-octyl phthalate (DOP)] and some other derivatives from PET bottles into their water and vinegar.

EXPERIMENTAL SECTION

Materials

- Eighteen Samples of bottled water and vinegar backed in polyethylene terephthalate bottles (PET) with a volume capacity of 1000 ml, were collected randomly from the local market at start time

(production date from 1-7 days).

- Stock standard solutions of phthalate derivatives (were purchased from Sigma).

Methods

Storage condition

The samples were divided into three groups according to the condition of storage (temperature and time).

Group 1: bottled water and vinegar stored at 4 ° C in the refrigerator (leaching measured at start time, for two and four months).

Group 2: bottled water and vinegar stored at 20 ° C in the furnace (leaching measured at start time, for two and four months).

Group 3 bottled water and vinegar stored at 40 ° C in the furnace (leaching measured at start time, for two and four months).

Preparation of plasticizers standard

Standard solutions (1.0 µg/µL); can be prepared from pure standard materials or purchased as certified solutions.

Preparation of samples

The volume of the sample, approximately 1 L, is serially extracted with methylene chloride

at a pH greater than 11 by using a separator funnel or a continuous extractor method 625 (EPA, 2007).

Determination of plasticizers (phthalate derivatives)

Phthalate compounds contents in bottled water and vinegar samples under investigation were determined using gas chromatography apparatus according to the method of EPA, (1984).

RESULTS AND DISCUSSION

The concept of the overall migration limit has mainly been used to control the total amount of substances migrating from plastics into food and determining the toxicological importance of the substances (Arvanitoyannis and Bosnea, 2004)

The migration process can be divided into four major steps: diffusion of chemical compounds through the polymers, desorption of the diffused molecules from the polymer surface, sorption of the compounds at the plastic–food interface, and desorption of the compounds in the food (Ferrara et al., 2001).

Migration of plasticizer compounds from PET bottles into

samples (collected from local markets) was studied and the obtained results are shown in table 1 and 2, figures (1, 2). The study observed the migration of derivatives PET to samples stored in PET bottles at different temperatures and periods.

When vinegar was stored at, the start time of 4 °C there was no contamination with plasticizers compounds from PET bottles. While at the same time at 20 °C PET derivatives such as DEHP and DBP appeared with concentrations of 25.1 and 3.5 µg/l but at 40 °C DEHP, BBP increased to 31.9 and 6.0 µg/l respectively, which showed in table 1 and represented in figure (1).

Migration of PET derivatives such as DEHP, DBP, and BBP increased at a high rate from start time to two months at 4 °C by 23.7, 10.72. When vinegar was stored at the start time of 4 °C there was no contamination with plasticizers compounds from PET bottles. While at the same time at 20 °C PET derivatives such as DEHP and DBP have appeared with concentrations of 25.1 and 3.5 µg/l but at 40 °C DEHP, BBP increased to 31.9 and 6.0 µg/l respectively, which showed in

table 1 and represented in figure (1).

Migration of PET derivatives such as DEHP, DBP, and BBP increased at a high rate from start time to two months at 4 °C by 23.7, 10.72, and 6.34 µg/l respectively while DMP, DEP, Azobenzene, and Phenanthrene have appeared in minor concentrations.

While at 20 °C, results showed that the migration of these components increased with increasing temperature and storage period. All derivatives of PET at 40 °C after two months were increased more than at 4 °C and 20 °C while fluorene and pyrene do not appear.

Regarding the same table and figure after four months of migration for all PET derivatives to vinegar stored in PET bottles noticed that at 40°C DEHP and DBP increased by increasing time to 37, 23.5µg/l respectively, DMP and DEP increased with small concentrations.

At 20 °C DEHP, DBP, and BBP also increased by increasing temperature and reaching a maximum concentration of 51.8, 36.3, and 15.4 µg/l at 40 °C respectively.

In addition, a rise in temperatures and storage time showed increases in levels of migration with linear relationship (R²) for DEHP ranging from (0.72-1.0), for DBP reached 1.0, and for BBP (0.8-0.9).

The results in the present study for vinegar as 3% acetic acid were counteractive to those reported by **Farhoodi et al., (2008)** who studied the interaction between PET bottles and a solution of 3% acetic acid as a simulation of acidic food. Over incubation time with different storage temperature and studied the migration of DEHP from PET bottles by incubation the solution in PET bottles for up to 120 days, either at 25°C or at 45°C. At the beginning of the trial, the amount of DEHP in PET bottles at different temperatures was below the limits of detection while this study reached a start time of 25.1 at 20 °C and 31.9µg/l at 40°C. These results were maybe because they used acetic acid as a food simulation, while in this study, food (vinegar) was utilized.

In addition, **Farhoodi et al., (2008)** disagree with the results obtained from this study they found that after incubation solution of 3% acetic acid, the amount of DEHP in

the solution incubated at 25°C for 25 days was 1.2 mg/L, whereas the amount of DEHP in the solution incubated at 45°C was 2.1 mg/L. The amount of DEHP in the solution brooded at 25°C had peaked at 1.4 mg/L, whereas the value of DEHP in the solution incubated at 45°C was 2.5 mg/L after a period (66 days), while the maximum concentration of DEHP in the present study was 51.8µg/l.

As the temperature increases, the diffusion of monomers and other compounds increased, and it can result in higher diffusion or rates of migration from packaging materials. The diffusion coefficient of packaging material components increased to 6-7fold when packages are exposed to extreme temperature fluctuations (for example, from freezer temperatures to cooking temperature.

The results obtained from this study were matching with those obtained by **Fasano et al., (2012)** who determined the migration of phthalates from a wide range of food-packaging materials to food simulants (3% acetic acid, distilled water, and 15% ethanol) and found higher

amounts of plasticizers released from PET bottles at 40°C.

The findings in a table (2) and Figure (2) displayed the migration of plasticizers compounds (DEHP, DBP, BBP, DEP, DMP, Azobenzene, Phenanthrene, Fluorene, and Naphthalene) to bottled water. When bottled water is stored for the first time at 4°C there is no contamination with plasticizers compounds from PET bottles except DEHP with concentration 3.3µg/l and DBP with concentration 0.078µg/l.

These scores were in harmony with those reported by **jeddi et al., (2014)** their conclusion is based on a comparison of the phthalates levels in bottled water packed in PET with bottled water packed in glass. The presence of higher levels of phthalates in PET bottled water indicates that PET is the main source of phthalates

In addition, table (2) illustrated that at the same time at 20°C the concentration of DEHP increased. At 40°C BBP appeared and DEHP concentration increased by increasing temperature. These results were conformity with those reported by **Jeddi et al., (2014)** who studied the concentrations of

Phthalates in bottled water when stored under different storage conditions and reported that concentrations of phthalate before storage (after production) were very low for DEHP and DBP while BBP was not found at detectable levels in PET-bottled water prior to storage.

Jeddi et al., (2014) also reported that concentrations of DBP and DEHP in all water samples classified by their storage conditions. While BBP was not detected at, low temperatures and interpreted, the presence of phthalates in PET bottled water at the start time to confirm that they might come from water treatment facilities, namely: pipes, storage tanks, and filtering systems.

The obtained results were inconsistent with **Al-saleh and Elkhatib, (2014)** who reported that bottled water stored at 4 °C contains higher levels of DMP, DEP, BBP, and DEHP than those stored at room temperature and outdoors. On the other hand, level of DMP, DEP, and BBP were significantly lower in bottled water stored at room temperature than those outdoor it seems that temperature and sunlight play a

role in the degradation of phthalate over time.

Regarding table (2) and Figure (2) after storage for two months at 4 °C DEHP and DBP showed increasing in concentrations with increasing storage time by 2.4, and 20 times respectively. Dinitrobenzene has appeared with small concentration. while at 20°C in the same time the DEHP increased 2.4 times, DBP increased by 10 times, BBP at the beginning of appearances at this condition, Azobenzene, and DMP appeared with small concentrations. DEHP, DBP, and BBP were increased by 2.4, 17.4, and 21 times respectively after two months at 20 °C. Azobenzene appeared by increasing the temperature to 40 °C. During the same period of storage, all target compounds increased in concentration by increasing temperature. This clearly appeared by linear relationship with $R^2 = 0.9$ for DEHP, ranged from (0.81-0.92), and BBP (0.75-0.92).

The obtained results go parallel with those obtained by **Casajuana and Lacorte, (2003)** who concluded that poor storage conditions 10 weeks outdoors at temperatures of up to 30°C)

increased the concentrations of DBP, BBP and DEHP from bottled water.

Migration of all derivatives PET to bottled water stored in PET bottles was noticed after four months Table (2) and Figure (2) showed increasing in concentration with increasing temperatures. DEHP represents the max concentration of PET derivative 62.5µg/l while DBP showed a high increase after four months by 11.3, 5.9, 3.81 times at (4, 20, and 40°C) respectively than that after two months.

The previous results were also in disagreement with the observation of **Amiridou and Voutsas, (2011)** who did not find a difference in the concentrations of phthalates (DEP, DBP, and DEHP) in water from PET bottles exposed to different storage conditions (start time, 15 and 30 days) at different temperatures(15 - 40°C).

In addition, disagreement with the observation of **Wittassek et al., (2011); Koch and Calafat, (2009)** who said that the exposure to DEP, DBP, BBP, and DEHP is significantly decreased over time, whereas the exposure to DiNP, DiDP, and DiBP is on a rising trend. The present results also

disagree with **Farhoodi et al., (2008)** detected DEHP in their samples when first tested, after 66 days, the concentration of DEHP in their sample reached 2.5 mg/L (i.e., 2500 µg/L) with incubation at 45°C. Likewise, **Biscardi et al., (2003)** reported similar concentrations of DEHP but after 9 months of incubation of water in PET bottles at room temperature.

The maximum permitted level for DEHP maximum contaminant level (MCL) is determined by EPA standards and WHO guidelines (6/8 µg/L respectively) (**FDA, 2011 and EPA, 2009**). The concentration of DEHP in this study exceeded the MCL determined by EPA standards and WHO guidelines by 10, 7.5 times. US EPA has suggested a maximum contaminant level of 10 µg/L for BBP in drinking water the values of BBP in this study exceeded the maximum contaminant level.

CONCLUSION:

The results showed that the highest migration level was DEHP after 4 months at 40°C reached 62.5 and 51.9 µg/l in bottled water and vinegar these values were 10 and 8.6 times higher than the upper limit for maximum contamination level (MCL) in drinking water as

regulated by U.S.EPA, which equal 6.0 µg/L for DEHP. Migration increased with increasing temperature and storage time with linearity reached one.

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Table (1): The level of migration of PET derivatives to vinegar Conc. 3% (mean pH = 2.8) over the storage periods (start time, after 2 and 4 months) at different temperatures at (4, 20 and 40°C).

PET derivatives µg/l	Temp.	Three periods/ months			R ²	Equation	At EXP (12 months) calculated
		start time	after2 months	after 4 months			
DEHP	4 °C	0	23.7	37.0	0.97	Y=18.5-16.7	205
	20 °C	25.1	27.0	43.24	0.83	Y=9.0x+13.7	122
	40 °C	31.9	38.32	51.85	0.72	Y=10.3+17.7	142
DBP	4c °C	0	10.72	23.54	1.0	Y=11.7x-12	128
	20 °C	3.5	24.97	28.71	0.86	Y=12.6x-6.1	145
	40 °C	6.0	18.74	36.37	0.9	Y=15.6x-8.0	180
BBP	4 °C	0	6.34	9.76	0.97	Y=4.8x-4.3	54
	20 °C	0	11.481	12.9	0.83	Y=6.4x-4.7	72
	40 °C	0	13.53	15.4	0.84	Y=7.7x-5.7	86
DEP	4°C	0	0.192	0.25			
	20°C	0	0.194	0.29			
	40 °C	0	0.236	0.33			
DMP	4 °C	0	0.145	0.29			
	20 °C	0	0.180	0.33			
	40 °C	0	0.311	0.36			
Naphthalen	4 °C	0	0	0			
	20 °C	0	0	0			
	40 °C	0	0.07	0.13			
Azobenzene	4 °C	0	0.289	0.3			
	20 °C	0	0.353	0.35			
	40 °C	0	0.389	0.52			
Phenanthrene	4 °C	0	0.247	0.25			
	20 °C	0	0.258	0.26			
	40 °C	0	0.293	0.33			

Hypothesis Migration of Plasticizers from Polyethylene Terephthalate Bottles to Vinegar and Water via different Temperature and Storage Periods

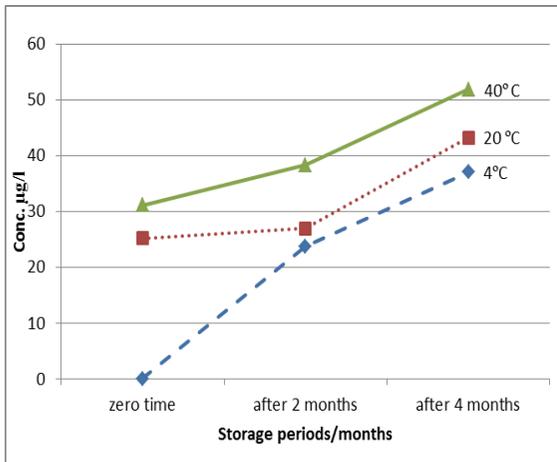
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Table (2): The level of migration of PET derivatives to bottled water (mean pH = 6.5) over the storage periods (start time, after 2 and 4 months) at different temperatures at (4, 20 and 40°C).

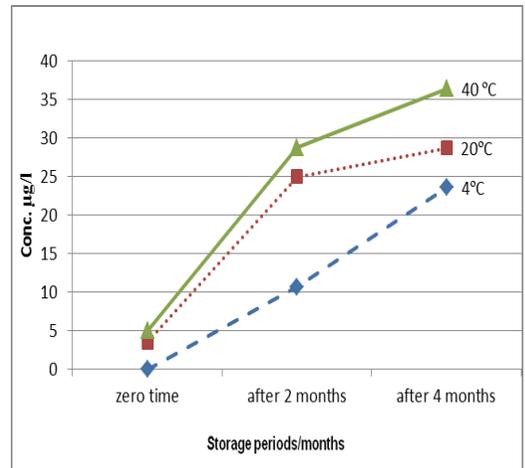
PET derivatives µg/l	Temp.	three periods/ months			R ²	Equation	At EXP. (12months) calculated
		start time	after 2 months	after 4 months			
DEHP	4 °C	3.3	8.08	27.69	0.9	12.2x-11	135
	20 °C	5.15	12.67	46	0.9	20.4x-19	225
	40 °C	6.11	14.68	62.52	0.9	28.2x-28	309
DBP	4 °C	0.08	1.6	18.12	0.81	9.024x-1	92
	20 °C	0.47	4.42	26	0.87	12.9x-15	13 9
	40 °C	0.8	10.75	40.99	0.92	20.1x-22	21 8
BBP	4 °C	0	0	1.458	0.75	1.73x-2	18
	20 °C	0	2.95	4.897	0.97	2.45x-2	27
	40 °C	0.22	4.75	16.50	0.94	8.14x-9	88
DEP	4 °C	0	0	0.21			
	20 °C	0	0.154	0.3			
	40 °C	0	0.35	0.5			
DMP	4 °C	0	0	0.155			
	20 °C	0	0.138	0.2			
	40 °C	0	0.302	0.335			
Naphthalene	4 °C	0	0	0			
	20 °C	0	0	0			
	40 °C	0	0.042	0.209			
Azobenzene	4 °C	0	0	0.404			
	20 °C	0	0.192	0.603			
	40 °C	0	0.21	1.134			
Phenanthrene	4 °C	0	0.2	0.344			
	20 °C	0	0.278	0.328			
	40 °C	0	0.2	0.344			

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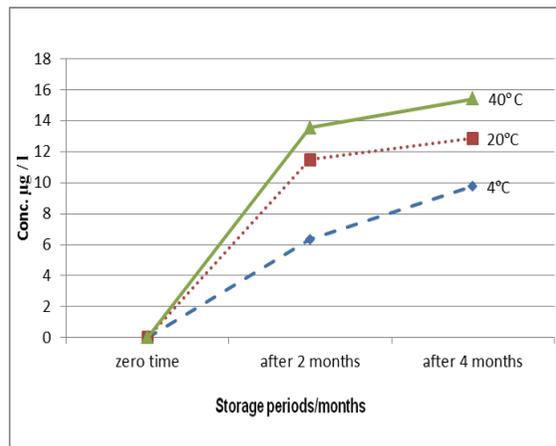
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a) DEHP



b) DBP

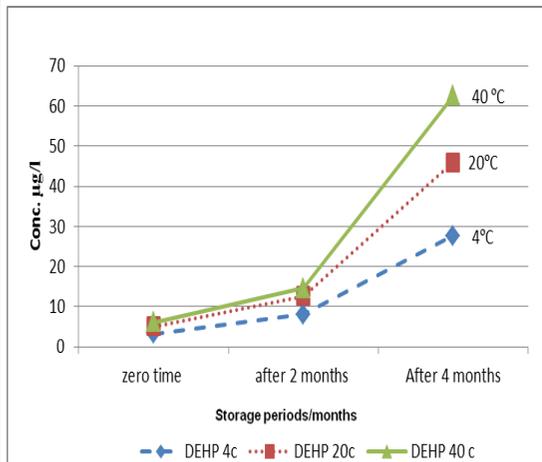


c) BBP

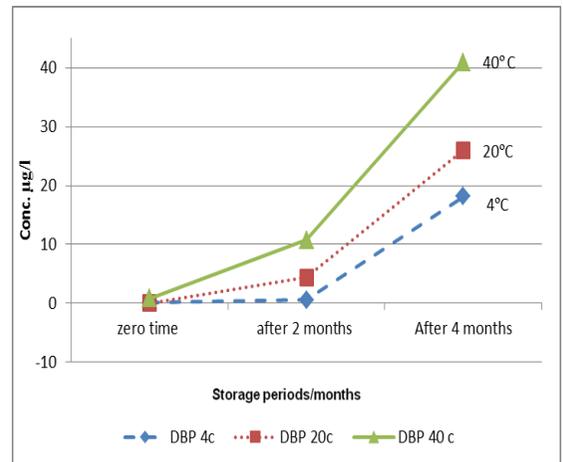
Figure (1) The level of migration of PET Derivatives to Vinegar Stored in PET Bottle

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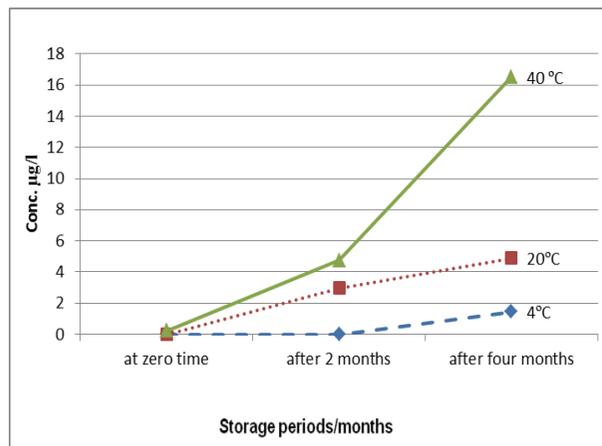
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a)DEHP



b) DBP



C)BBP

Figure (2) The level of migration Of PET Derivatives To Bottled Water Stored In PET Bottle

هجرة الملدنات من زجاجات البولي إيثيلين ثلاثي الفثالات إلى الخل والمياه عبر فترات تخزين ودرجات الحرارة المختلفة

سهام أديب عرابي ١، طه عبد العظيم عبد الرازق ٢ و هناع حسين السيد ١*

١- قسم كيمياء الاغذية والتمثيل الغذائي - المعهد القومي للتغذية - القاهرة - مصر
٢- قسم العلوم الاساسية البيئية - كلية الدراسات والبحوث البيئية - جامعة عين شمس القاهرة - مصر

الملخص العربي

تستخدم الملدنات (ثنائي إيثيل الفثالات (DEP) وثنائي بيوتيل الفثالات (DBP) وبنزويل بيوتيل فثالات (BBP) وثنائي إيثيل هيكسيل فثالات (DEHP) على نطاق واسع في صناعة البلاستيك. لا ترتبط الفثالات تساهمياً بالمصفوفة البلاستيكية ويمكنها تتسرب إلى المواد الغذائية أثناء التعبئة، وقد بحثت هذه الدراسة في انتقال مشتقات الفثالات من عبوات البولي إيثيلين تيريفثاليت (PET) إلى المياه والخل المعبأة بها تحت ظروف تخزين مختلفة. تم استخدام كروماتوغرافيا الغاز (GC) مع نظام قياس الطيف الكتلي (MS) وأظهرت النتائج أن أعلى مستوى انتقال كان مستويات DEHP بعد ٤ أشهر عند ٤٠ درجة مئوية وصلت إلى ٦٢,٥ و ٥١,٩ ميكروغرام / لتر في المياه المعبأة والخل، كانت هذه القيم أعلى ١٠ و ٨ و ٦٥ مرة من الحد الأقصى لمستوى التلوث (MCL) في مياه الشرب وفقاً EPA-US، والتي تساوي ٦,٠ ميكروغرام / لتر. DEHP.

الكلمات المفتاحية: الملدنات، الهجرة، التخزين، درجات الحرارة