Diffusion and adsorption of BaP from vegetable oils onto polyethylene terephtalate and low density polyethylene package

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Summary

Sunflower and rapeseed oils were spiked with benzo[*a*]pyrene (BaP) at the level of 37.1 and 29.4 μg.kg^{-1}, respectively. Then, the oils were filled into polyethylene terephtalate (PET) and low density polyethylene (LDPE) cylindrical shape receptacles and BaP concentrations were followed within 49 h using HPLC with fluorescence detection. It was found that the concentration in the oils in contact with PET were decreased considerably (sunflower oil 25.9 μg.kg^{-1} and rapeseed oil 22. 8 μg.kg^{-1}) due to sorption of BaP onto PET, while no decrease in concentration of BaP was observed in the oils in contact with PE. It was possible to describe the rate of BaP removal by kinetic equation and calculate diffusion coefficients. The results showed that PET is able to decrease BaP concentration in both vegetable oils and the decrease is due to monolayer sorption processes taking place on the PET surface. As found, LDPE is inappropriate material for BaP removal from sunflower and rapeseed oils, because BaP concentration in the oils remained at a constant level during the whole experiment.

Keywords

polycyclic aromatic hydrocarbons; benzo[*a*]pyrene; sunflower oil; rapeseed oil; polyethylene terephtalate; polyethylene; adsorption

Polycyclic aromatic hydrocarbons (PAHs) belong to hazardous contaminants due to their known or suspected carcinogenicity and/or mutagenicity. In general, PAHs are formed by incomplete combustion of fossil fuels and other forms of organic matter. For these reasons they are being found in all parts of the environment, including foods [1]. Moreover, PAHs are also formed at thermal processes during of food production such as baking, grilling, roasting, frying, and smoking [2, 3]. With regard to the harmful effects of PAHs on living organisms, there are trends to enact maximum limits of these compounds in various foods to protect consumers against harmful effects of these compounds. To simplify difficulties associated with a great variability of PAH composition, benzo[*a*]pyrene (BaP) has been accepted, in general, as the indicator of total PAH presence in foods regardless of the fact that BaP constitutes only between 1–20 % of the total carcinogenic PAHs [4]. Relating to PAH legal limits, the situation in the EU has been changed considerably due to the adoption of the regulation 208/2005 limiting the BaP concentration to the level of 2 μg.kg^{-1} in oils and fats intended for direct human consumption or use as an ingredient in foods [5]. This regulation has come into force from 28th February 2005 and started to be applied from 1st April 2005. Apart from this, EC has also adopted the directive 2005/10/EC laying down the sampling methods and the methods of analysis for the official control of BaP levels in foodstuffs [6] and the recommendation 2005/108/EC on further investigation into the levels of PAHs in certain foods as follows: benzo[*a*]anthracene, benzo[*b*]fluoranthene, benzo[*j*]fluoranthen, benzo[*k*]-fluoranthene, benzo[*g,h,i*]perylen, BaP, chrysene, cyclopenta[*c,d*]pyrene, dibenzo[*a,h*]anthracene, dibenzo[*a,e*]pyrene, dibenzo[*a,c*]anthracene, dibenzo[*a,i*]pyrene, dibenzo[*a,l*]pyrene, indeno[*1,2,3-c,d*]pyrene, and 5-methylchrysene [7]. Fats and oils belong to the most important sources of

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PAH intake due to their extensive consumption as well as PAH concentrations which can sometimes reach high levels. For example, MORET *et al.* [8] analysed 51 samples of olive oils and determined the total PAH concentrations from 2.94 μg.kg^{-1} to 143.12 μg.kg^{-1}. HOPIA *et al.* [9] found various PAHs in Finnish butters, margarines and vegetable oils, and some raw vegetable oil materials, when total PAH concentrations in 25 samples varied from 0.17 μg.kg^{-1} (corn oil) to 4600 μg.kg^{-1} (crude coconut oil). It was concluded that the enormous PAH concentration in coconut oil could be brought about by direct drying of copra with smoke. BARRANCO *et al.* [10] pointed out technological procedures of deodorization and bleaching, which are able to decrease PAH concentrations considerably in oils during production. An over limited concentration of BaP in Spanish olive oils was the reason to withdraw them from central European markets [11]. Polymers could play an important protective role against PAHs exposition with regard to high affinity of PAHs to some plastic materials [12]. For example, PAH concentrations were reduced effectively in a liquid smoke flavour by two orders of magnitude during 14 days of storage in LDPE flasks [13]. It was found that the rate-limiting step of the PAH sorption from liquid into LDPE is diffusion in liquid media [14]. PAHs are primarily adsorbed on the LDPE surface with subsequent diffusion into polymer bulk [15]. The most intense processes take place within the first 24 h, when the polarity and viscosity of liquid media play an important role. During this period, the PAH concentration was lowered in three different liquid systems by more than 50 % from the initial concentration of 50 μg.kg^{-1} for each tested compound [16]. The ability of PET to decrease PAH concentrations in polar and non-polar liquid media has already been unambiguously proven [11]. However, the extent of removal processes is strongly affected by the presence of other PAH compounds as well as compounds such as vitamins, sterols, waxes present in rapeseed oil [17]. On the basis of the current knowledge mentioned above, the aim of this work was to study effects of PET and LDPE on BaP contained in sunflower and rapeseed oil.

MATERIALS AND METHODS

Sunflower and rapeseed oil

Commercially available sunflower and rapeseed oils were produced by Palma-Tumys (Bratislava, Slovakia) and purchased in a local market in Bratislava. The oils were packed in PET bottles with volume of 4 l.

PET receptacles

In the experiment, pre-bubbled PET receptacles of cylindrical shape with i. d. of 21.4 mm and height 150 mm were used. The receptacles were provided by Palma-Tumys. The company uses them for oil and fruit syrup packaging after blowing to volume of 2 l.

LDPE receptacles

LDPE cylindrical shape receptacles with i. d. of 32 mm and height of 80 mm were supplied by Čechvalab (Bratislava, Slovakia).

BaP

BaP was of analytical grade, purchased from Supelco (Bellefonte, PA, USA) in a solid state. A solution for spiking was prepared by dissolving BaP in acetonitrile to get the initial concentration of 500 mg.l^{-1}.

Solvents

Acetonitrile was of HPLC grade (Merck, Darmstadt, Germany), methanol, and hexane for analysis (Slavus, Bratislava, Slovakia). The solvents were rectified just before use in a distillation apparatus.

Other chemicals and materials

Anhydrous Na_2SO_4 and alumina were purchased from Merck.

Experiment

First of all, the oils were analysed for the presence of BaP to know initial - "natural" BaP concentration in the oils. Subsequently, 100 g of the oil was spiked with BaP solution in 2 l volume glass flask and the solvent left to evaporate spontaneously. To accelerate the evaporation, the oil was stirred up occasionally. Then, roughly 900 g of the oil was added and the content of the flask was stirred thoroughly. At this stage, a sample of spiked oil was taken to determine the initial BaP concentration. Then, the PET and LDPE receptacles were filled with the spiked oils and placed into a polystyrene box to protect them from light and to keep at a constant temperature. The samples for analysis were taken after 1; 3; 5; 7; 11; 24; 49 h. To maintain the same static conditions and sampling during the experiments, a new set of receptacles was taken for each analysis.

Sample preparation

Sample preparation was performed according to ISO 15302 [18] as follows: 2 g of oil was weighed to the nearest 0.001 g into a 10 ml graduated flask, dissolved in hexane and diluted to the mark. Then a chromatography column filled with hexane, and 22 g of alumina transferred immediately to the col-

umn, and anhydrous Na_2SO_4 was added to the top of the column to form a layer about 30 mm thick. Then hexane was let to fall to the level of the top of Na_2SO_4 layer, and 2 ml of the graduated flask content was applied onto the column. The column was eluted with hexane with a flow of about 1 ml.min^{-1}, discarding the first 20 ml of eluate and then collecting 60 ml of eluate in a 100 ml round-bottomed flask. The eluate was evaporated to about 0.5 ml, and transferred into a vial. Evaporation continued under nitrogen until nearly dry. Round-bottomed flask was rinsed with about 1 ml of hexane twice and transferred quantitatively to the mini-vial, and continued in evaporation under nitrogen. The evaporation was carried out until dry, the residuum dissolved in methanol and analyzed by HPLC.

HPLC analysis

Analysis were performed on liquid chromatograph Agilent Technologies 1100 Series (Halbron, Germany) consisting of a quaternary pump, micro vacuum degasser, autosampler, and fluorescence detector, which operated at 300 nm excitation and 410 nm emission wavelengths. Separations were carried out at ambient temperature on column LiChrolut (Merck) 25 cm x 0.4 cm i. d. packed with Lichrospher PAH (particle diameter 5 μm), when pre-column LiChroCart (4 cm x 0.4 cm) with the same particle diameter was also used. The flow rate of mobile phase was 1 ml.min^{-1}. For determination, a gradient elution was used as follows: A - deionized water, B - acetonitrile. Gradient programme: from 0 to 2 min elution with 30 % A and 70 % B, then from 2 to 5 min from 70 % B to 100 % B linearly, then elution from 5 to 10 min isocratically, then from 10 to 15 min from 100 % B to 70 % B linearly. Equilibration time between each run was 5 min. Samples were applied using an autosampler needle with 20 μl volume. All analyses were carried out in duplicate with an average relative standard deviation of 8.4 %.

RESULTS AND DISCUSSION

At first, the experiment was carried out with sunflower oil to be filled in PET receptacles. The experimentally obtained dependence of BaP concentration *vs*. time was used for the calculation of diffusion coefficient *D* using a kinetic equation (1), which has been derived for diffusion of PAHs in non-stirred liquids placed into cylindrically shaped plastic receptacles [11]

$$c_t = c_\infty + (c_0 - c_\infty)\sum_{n=1}^{\infty}\frac{4}{a^2\alpha_n^2}\exp\left[-D\alpha_n^2 t\right] \qquad (1)$$

where C_0 is the initial concentration of BaP in oil, C_t is the concentration of BaP, which remained in the oil at time t, and C_∞ is the concentration of BaP corresponding to infinite time, a is the radius of the cylinder, and α_n are the roots of the equation:

$$J_0(a.\alpha_n) = 0 \qquad (2)$$

where J_0 is the zero-order first-kind Bessel function.

D was calculated by the non-linear least squares method by minimizing the sum of squares of differences between the BaP concentrations measured experimentally and those calculated by equation (1) and expresses a rate of BaP diffusion in the oil. As follows from Fig. 1, BaP concentration in the oil began to decrease immediately after filling the PET receptacles to reach the equilibrium concentration after 24th h of experiment. The same situation was observed in the rapeseed oil, when a part of BaP was adsorbed onto PET surface, a part remained in the oil and the equilibrium concentration was established after 24th h of experiment (Fig. 2). Values of BaP diffusion coefficients, calculated from equation (1) for both oils are almost identical what indicates to the same physicochemical processes taking place in liquid media (Tab. 1). Very interesting is comparison of "total BaP area"

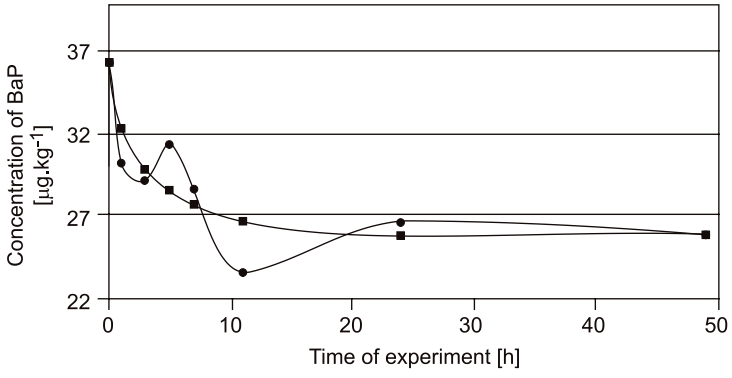

Fig. 1. Changes in BaP concentration in sunflower oil stored in PET receptacles. ● - experimentally obtained data, ■ - calculated data using kinetic equation of adsorption (1).

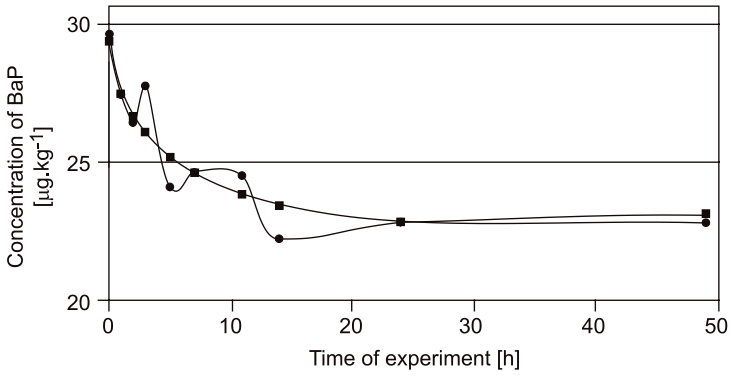

Fig. 2. Changes in BaP concentration in rapeseed oil stored in PET receptacles. ● - experimentally obtained data, ■ - calculated data using kinetic equation of adsorption (1).

Tab. 1. Parameters calculated from experimentally measured values.

	Diffusion coefficient *D* [$cm^2.h^{-1}$]	PET area occupied by BaP molecules [%]	Temperature during experiment [°C]
sunflower oil	3.9×10^{-2}	37	23.2 ± 0.3
rapeseed oil	2.5×10^{-2}	21	18.3 ± 0.4

to contact area at the boundary PET-vegetable oil. Size of BaP molecule was calculated using Savol program (Tripos, St. Louis, Missouri, USA) and is equal to 2.89 nm^2. Number of BaP molecules adsorbed on PET surface was calculated from differences between initial and equilibrium BaP concentrations in the oils. Results showed that the area of BaP adsorbed onto PET from sunflower oil is equal to 37 % of total PET area, while for BaP adsorbed in rapeseed oil the area is 21 %. These values indicate that the BaP removal from vegetable oils could be classified as monomolecular adsorption on PET surface. Because the removal of BaP from oils packed into PET is limited by surface adsorption and the diffusion of BaP into LDPE bulk has been already proven as more effective material, LDPE was studied for BaP removal from the vegetable oils. However, BaP concentration in both oils remained at a constant level, as shown in Fig. 3. This observation is really surprising with regard to previous findings, because LDPE functioned effectively and it was able to decrease PAH concentration by one [16] or two orders [14] in liquid media. However, one explanation could be based on the fact, that LDPE was used effectively for removal of PAHs from polar, or semi-polar liquid media. Oils are matrices which are either non-polar or contain double bonds able to interact via π-electron pairs with delocalised π-electrons of PAHs. As these interactions are probably stronger in comparison with interactions of PAHs with LDPE (which is free of π-electron pairs), they are able to hinder from PAH migration into LDPE and maintain the BaP concentration at a constant level. As can be seen from Figures 1 and 2, the dependencies of BaP concentrations on time exhibit oscillations, especially at the beginning of the adsorption processes. These oscillations occur at the same time not only for both oils, but also have already been observed in paraffin oil, where the concentration was measured directly

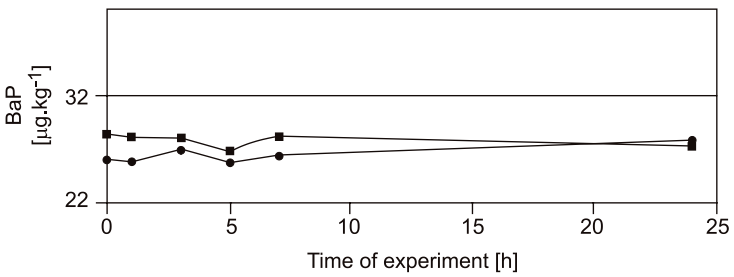

Fig. 3. BaP concentration in sunflower (■) and rapeseed (●) oils stored in PE receptacles.

without sample preparation procedures to exclude possible experimental errors [17]. It indicates that the adsorption processes in the systems PET-vegetable oils are complicated, or even combined with another, at this time, unknown processes.

CONCLUSIONS

The results and findings of this work lead to the following conclusions:

1. The BaP concentration in the vegetable oils filled into PET can be decreased due to interaction between BaP contained in the oils as a liquid phase and PET as a solid phase.
2. It seems that these interactions could be classified as monomolecular adsorption of BaP on PET surface.
3. It was found that LDPE is not appropriate material for elimination of PAHs from a nonpolar matrix, e. g. vegetable oils.
4. The dependencies of BaP concentrations on time exhibit oscillations, especially at the beginning of interaction - the reasons of their existence are unknown so far.
5. This knowledge might be used in industrial production of vegetable oils inserting additional operation of PAH sorption on surface PET particles just after bleaching procedures to remove residual PAHs, and in such way to protect human organism against exposure to these carcinogenic compounds.

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