Biomonitoring of polyaromatic hydrocarbon accumulation in rural gardens using lettuce plants

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Abstract

Purpose Air pollution in settlements is one of the most important factors affecting human health. While urban gardening and the consumption of home-grown vegetables have become more and more popular, several studies establish the risk of consuming vegetables exposed to toxic air pollutants, especially polyaromatic hydrocarbons (PAHs). However, these studies mostly concern the risk in larger cities or in industrial areas while much less information is available in rural environments.

Methods In our study, small settlements were selected for a pot accumulation study. Lettuce (*Lactuca sativa*) plants were exposed for a 2-month period; PAH concentrations were determined both from the vegetable samples and pot soils.

Results In the lettuce samples, the concentration of total PAHs was in the range of 9.1 (Litér) and 185 µg/kg dry wt (Hajmáskér), in the soil samples in the range of 31.6 (Litér) and 595.1 µg/kg wt (Hárskút). Four ring PAHs showed the highest tendency to accumulate in the majority of soil samples.

Conclusion PAH concentrations in some of the sampled villages were comparable to results reported from relatively polluted regions in the world. Source appointment revealed that biomass burning and fossil fuel usage were the major sources of PAHs in rural environment; in addition, the study highlighted how important it could be to analyze individual pollution sources.

Keywords Polycyclic aromatic hydrocarbons · Kitchen gardens · *Lactuca sativa* · Bioaccumulation · Soil

1 Introduction

Polycyclic aromatic hydrocarbons (PAHs) in settlements are mainly derived from incomplete combustion in motor vehicles; another major source is domestic heating (Szabó Nagy and Szabó 2017). A wide range of PAHs has proven highly carcinogenic or mutagenic: for example, the so-called Car-PAHs (Srogi 2007), or the more extended list of 16 priority

PAHs identified by the US Environmental Protection Agency (EPA) in the 1970s.

In the air, PAHs occur in the gaseous phase (mainly PAHs with higher vapor pressure) and bound to particles (mainly PAHs with lower vapor pressure). Plants are exposed to both phases. Moreover, atmospheric PAHs are easily transported to the soil, providing an additional exposure route for plants. In general, possible uptake mechanisms can be summarized as follows: gas exchange; particle deposition (from air) and uptake (from soil) (Kulhánek et al. 2005).

Vapor phase PAHs might diffuse through the wax and the cuticular membrane into the interior of the leaves but they can also penetrate directly via the stomata (Lehndorff and Schwark 2004). Particle-bound PAHs were also found in the cuticle waxes (Kaupp et al. 2000). Yang et al. (2017a) reported that the transportation of PAHs from foliar deposition to the cuticular wax could be the primary pathway of leaf accumulation. Atmospheric uptake of halogenated PAHs in leafy vegetables was also demonstrated (Wang et al. 2018).

However, comparing uptake from soil and from the atmosphere, atmospheric exposure is considered as the dominant

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source of contamination (Phillips 1999). Concentration of PAHs in the aerial parts of plants are generally considerably higher than in the roots (Zhang et al. 2020). Jia et al. (2019) experimentally demonstrated that approximately 90% of total PAH absorption in vegetables came via airshoot absorption.

PAHs are easily accumulated in vegetables (e.g., Khillare et al. 2012; Abou-Arab et al. 2014; Xiong et al. 2017). It is assumed that crops cultivated in urban areas are generally exposed to a higher level of pollutants compared to crops from rural sites (Säumel et al. 2012). While there are studies available on PAH accumulation in urban community gardens (Amato-Lourenco et al. 2017), rural gardens have been scarcely investigated, though so-called organically farmed vegetables were frequently found to accumulate organic contaminants, including PAHs (Zohair et al. 2006). Our aim was to assess the level of air pollution via bioaccumulation studies in small-medium-sized villages in Veszprém County, Hungary.

For pot experiments, lettuce (*Lactuca sativa* L., family Asteraceae) was selected. *L. sativa* is a widely used test species in bioaccumulation studies, due to high foliar surface and thin cuticula (Schreck et al. 2012, 2013). It generally shows higher bioaccumulation capacity in comparative studies (e.g.,

Li et al. 2015; Mombo et al. 2016). Also, lettuce is a very popular vegetable in Hungarian kitchen gardens; the amount of home-grown lettuce was 608 tons in 2017 according to the National Statistical Agency (KSH 2018). Several varieties are in use, some of them are so early that the vegetation period might be partially overlapping with the heating season.

2 Material and Methods

2.1 Locations

Seven small/medium-sized villages were selected for the study. All of them are located in Veszprém County, Transdanubian Region, Hungary (Fig. 1). Number of inhabitants, size, and location are given in Table 1. In Eplény, 2 sampling spots were used. As a main road cuts through this village, gardens being nearby the road might be highly affected. As such, one sampling spot was located very close to the road, and the other in the same property, approximately 80 m away from the road.

Fig. 1 Location of the sampling sites. 1, Nagyvázsony; 2, Pécsely; 3, Tihany; 4, Hárskút; 5, Eplény1; 6, Eplény2; 7, Hajmáskér; 8, Litér

Table 1 Description of the selected villages

Site no.	Name of the village	No. of inhabitants	GPS	Area (km ²)
1	Nagyvázsony	1820	46° 58' 39.9" N 17° 41' 44.3" E	76.29
2	Pécsely	588	46° 57' 13.2" N 17° 46' 53.2" E	20.01
3	Tihany	1346	46° 54' 46.2" N 17° 53' 06.9" E	27.33
4	Hárskút	674	47° 11' 16.4" N 17° 48' 24.9" E	34.46
5–6	Eplény	514	47° 12' 36.3" N 17° 54' 49.6" E	8.28
7	Hajmáskér	3090	47° 08' 40.4" N 18° 01' 12.6" E	38.14
8	Litér	2241	47° 05' 48.0" N 18° 00' 24.8" E	12.83

2.2 Exposure experiment and sample collection

For the test, the variety “Május királya” (King of May) was used. It was selected as it is one of the most popular varieties cultivated in home gardens in Hungary. Seeds were purchased from Garafarm Ltd. Lettuce seedlings were grown for 40 days in a glasshouse. Five seedlings were placed in a plastic pot (diameter 31 cm). Five replicates (5 pots) were used at each site. Exposure took 2 month, between 29.03 and 31.05. During the study period, neither additional fertilizers nor pesticides were used. After the exposure, pots were collected and taken to the laboratory where the lettuce plants were washed with ionic load-free water and immediately frozen (− 20 °C) until analysis.

In the pots, commercial soil (pH, 6.8 ± 0.5 ; N (m/m%), min 0.3; P₂O₅ (m/m%), min 0.1; K₂O (m/m%), min 0.3) was used. The soil was analyzed prior to the experiment and concentration of PAHs was under the detection limit. In general, PAHs tend to accumulate in soils due to their strong hydrophobicity and resistance to degradation; it was found that approximately 90% of total PAHs retain in surface soils (Wild and Jones 1995). As such, at the end of exposure, the upper 5 cm layer of the soil was collected from each pot and the composite sample was frozen (− 20 °C) until analysis.

2.3 Analytical measurements

Ten grams of plant sample was grinded with 10-g anhydrous sodium sulfate in a ceramic mortar. The extraction was repeated 3 times with 20-ml n-hexane in ultrasonic extractor for 20 min. Prior to extraction, 10-ml acetone was added and the samples were spiked with 100 µl of 0.01 µg/ml deuterated PAH surrogate mixture (naphthalene-d₈, acenaphthene-d₁₀, phenanthrene-d₁₀, chrysene-d₁₂, benzo(a)pyrene-d₁₂, and perylene-d₁₂). The extract was concentrated with dry nitrogen stream to a 1 ml, and an additional solid-phase silica gel and alumina oxide sample clean-up was performed. The sample was passed through a 30-cm long, 0.5-cm width glass column containing 3 g of silica gel on the bottom of the column, and 3 g of aluminum oxide on the top, and it was eluted with 20-

ml methylene chloride. The column was washed with 20-ml n-hexane. The clean sample was concentrated with dry nitrogen stream to 1 ml. Before the measurement 100 µl of 0.01 µg/ml, internal standard mixture (2-floro-biphenyl, and p-terphenyl-d₁₄) was added (final concentration was 100 µg/kg plant dry wt). The plant samples were analyzed by Agilent 6890GC 5973E MSD GC-MS based on MSZ (Hungarian Standard) EN 15527:2009. Under the conditions specified in the standard, limit of detection of 0.1 µg/kg for each individual PAH can be achieved.

For measuring PAH content of the soil samples, the MSZ (Hungarian Standard) 21470-84:2002: Environmental protection, Testing of soils. Part 84: Determination of polycyclic aromatic hydrocarbons (PAH) content was followed. Under the conditions specified in the standard, typical limit of detection is 0.1 µg/kg soil sample. Gas chromatographic-mass spectrometric method Hungarian standard was followed. Ten grams of soil sample was grinded and extracted with 10-ml acetone and 10-ml n-hexane; the extraction was repeated two times; in the second and the third extraction, only 10 ml of hexane was used. Prior to extraction, the samples were spiked with 50-µl PAH surrogate mixture (deuterated, naphthalene D₈, acenaphthene D₁₀, phenanthrene D₁₀, chrysene D₁₂, benzpyrene D₁₂, perylene D₁₂) the final concentration was 100 µg/kg soil. The extract was dehydrated with sodium-sulfate and concentrated with dry nitrogen stream at 40 °C to a 1 ml; an additional solid-phase silica gel and alumina oxide sample clean-up was performed. The column was washed with 20-ml hexane and the extract was eluted with 20-ml dichloromethane. This extract was concentrated with dry nitrogen stream at 40 °C to a 1-ml, and the dissolvent was change to hexane. 100 µl of 0.01 µg/ml. Internal standard mixture (2-floro-biphenyl, and p-terphenyl-d₁₄) was added (final concentration was 100 µg/kg soil dry wt).

The GC-MS analysis was performed with an HP-6890 gas chromatograph; it was coupled to an HP-5973 (Agilent Technologies, Palo-Alto, USA) quadrupole mass spectrometer (low-resolution single MS). Injector and transfer-line temperatures were 320 °C and 250 °C,

respectively, and source and analyzer temperatures were 280 °C and 150 °C, respectively. A glass insert, 4 mm i.d., loosely filled with silanized glass wool was used in the split/splitless GC injector (320 °C, purge splitless 1.5 min). The GC column was 30 m × 0.25 mm i.d., film thickness 0.25 µm, ZB-Semivolatiles (Phenomenex). The GC oven temperature was maintained at 40 °C for 3 min after injection then programmed at 40 °C min⁻¹, 40 to 80 °C for 0.5 min, and then at 15 °C min⁻¹ to 240 °C which was maintained for 8 min and then at 15 °C min⁻¹ to 310 °C and which was maintained for 8 min. Helium (N55) was used as carrier gas at 1.2 mL min⁻¹.

The acquisition mode was SIM (single ion monitoring). Electron ionization was used with energy of 70 eV.

Analytical determinations were performed in the testing laboratory at the Laboratory of the ELGOSCAR-2000 Environmental Technology and Water Management Ltd. accredited by the National Accreditation Authority, registration number NAH-1-1278/2015.

For quality control, the glassware was thoroughly cleaned before use, washing with non-ionic detergent and rinsing with ultrapure water. Deuterated PAH surrogate mixture containing Naphthalene-d8, Acenaphthene-d10, Phenanthrene-d10, Chrysene-d12 Benzo(a)pyrene-d12, and Perylene-d12 was obtained from Restek Corporation (110 Benner Circle, Bellefonte, PA 16823, USA). Linearity of the calibration curve was checked in accordance with the accreditation standard.

3 Results and discussion

3.1 Accumulation in test plants

The 19 PAHs analyzed in this study included the following: naphthalene (Nap), 2-methyl-naphthalene (Methy-Nap), 1-methyl-naphthalene (Me-Nap), acenaphthylene (Acy), acenaphthene (Ace), fluorene (Flo), phenanthrene (Phe), anthracene (Ant), fluoranthene (Flt), pyrene (Pyr), benz(a)anthracene (BaA), chrysene (Cry), benzo[b]fluoranthene (BbF), benzo[k]fluoranthene (BkF), benzo[a]pyrene (BaP), benzo(e)pyrene (BeP), indeno[1,2,3-cd]pyrene (Ind), dibenz[a,h]anthracene (DahA), and benzo[g,h,i]perylene (BghiP). Their basic physico-chemical characteristic parameters are given in Table 2.

Table 3 summarizes the detect rate of analytically measured PAHs in both the lettuce and soil samples. In lettuce leaves, detect rate of only 4 PAHs was 100%: Nap, Methy-Nap, Phe, Flt while Acy, Ace, Ant, and DahA had a detection rate of 0.

Concentrations of all PAHs in the lettuce leaves are given in Table 4. Figure 2 shows the total amount of different molecular weight PAHs on the sampling spots, while Fig. 3 shows their percentage contribution. In all sites, the low molecular weight (LMW) PAH compounds were predominant in exposed lettuce plants (Fig. 2), which is in consistency with other studies (Lei et al. 2011; Wang et al. 2017;

Table 2 Basic physico-chemical characteristics of PAHs

PAHs	Abbreviation	Formular	Number of rings	Molecular weight (g)	Water solubility (mg/l) at 25 °C
Naphthalene	Nap	C ₁₀ H ₈	2	128.18	31
2-methyl-naphthalene	Methy-Nap	C ₁₁ H ₁₀	2	142.2	24.6
1-methyl-naphthalene	Me-Nap	C ₁₁ H ₁₀	2	142.2	28.5
Acenaphthylene	Acy	C ₁₂ H ₈	3	152.2	16
Acenaphthene	Ace	C ₁₂ H ₁₀	3	154.2	3.8
Fluorene	Flo	C ₁₃ H ₁₀	3	166.23	1.9
Phenanthrene	Phe	C ₁₄ H ₁₀	3	178.24	1.1
Anthracene	Ant	C ₁₄ H ₁₀	3	178.24	0.04
Fluoranthene	Flt	C ₁₆ H ₁₀	4	202.26	0.2
Pyrene	Pyr	C ₁₆ H ₁₀	4	202.26	0.13
Benzantracene	BaA	C ₁₈ H ₁₂	4	228.3	0.011
Chrysene	Cry	C ₁₈ H ₁₂	4	228.3	0.0019
Benzo(b)fluoranthene	BbF	C ₂₀ H ₁₂	5	252.32	0.0015
Benzo(k)fluoranthene	BkF	C ₂₀ H ₁₂	5	252.32	0.0008
Benzo(e)pyrene	BeP	C ₂₀ H ₁₂	5	252.32	0.007
Benzo(a)pyrene	BaP	C ₂₀ H ₁₂	5	252.32	0.0015
Dibenzo[a,h]anthracene	DahA	C ₂₂ H ₁₄	5	278.36	0.005
Indeno1.2.3CD-Pyrene	Ind	C ₂₂ H ₁₂	6	276.34	0.00019
Benzo(g,h,i)perylene	BghiP	C ₂₂ H ₁₂	6	276.34	0.00014

Table 3. Detect rates of individual PAHs in lettuce and soil samples

PAH compounds	Total soil sample (<i>n</i> = 8)		Total lettuce sample (<i>n</i> = 8)	
	Mean (range)	Detect rate	Mean (range)	Detect rate
Naphthalene	29.9 (0–154)	87.5	34.3 (3.1–155)	100
2-methyl-naphthalene	2.29 (1.2–5)	100	3.2 (1.5–6)	100
1-methyl-naphthalene	0.9 (0–3.1)	50	1.3 (0–2.8)	62.5
Acenaphthylene	0 (0)	0	0 (0)	0
Acenaphthene	0.6 (0–4.8)	12.5	0 (0)	0
Fluorene	0.8 (0–4)	37.5	1.0 (0–1.9)	75
Phenanthrene	12.3 (2.3–56.5)	100	6.8 (2.2–11.6)	100
Anthracene	2.7 (0–21.8)	12.5	0 (0)	0
Fluoranthene	20.3 (4.7–107)	100	3.7 (1.2–11.2)	100
Pyrene	15.6 (3.3–86.2)	100	2.4 (0–7.3)	87.5
Benzanthracene	10.4 (1.4–68.2)	100	1.2 (0–3.9)	62.5
Chrysene	7.9 (1.5–43.3)	100	1.5 (0–4)	75
Benzo(b)fluoranthene	11.5 (4.2–57.9)	100	1.9 (0–5.1)	75
Benzo(k)fluoranthene	4.3 (1.1–24.3)	100	0.4 (0–1.6)	25
Benzo(e)pyrene	5.9 (0–38.7)	100	0.8 (0–1.9)	62.5
Benzo(a)pyrene	5.1 (1.5–24.4)	100	0.8 (0–2.3)	50
Dibenzo[a,h]anthracene	0.7 (0–5.3)	12.5	0 (0)	0
Indeno1.2.3CD-Pyrene	3.7 (1.1–17.7)	100	0.4 (0–1.6)	25
Benzo(g,h,i)perylene	3.2 (1.2–12.7)	100	0.4 (0–1.9)	25

Table 4 Concentration of PAHs in lettuce samples. Priority PAHs are given in bold

PAH compounds	PAHs concentration (µg/kg dry wt) in lettuce leaves							
	Pécsely	Nagyvázsony	Eplény2	Eplény1	Tihany	Litér	Hárskút	Hajmáskér
Naphthalene	15.4	9.76	3.94	41.9	12.7	3.1	32.7	155
2-methyl-naphthalene	1.6	5.6	1.5	2	2.5	1.5	5.2	6
1-methyl-naphthalene	1.6	2.2	< 0.01	< 0.01	1.3	< 0.01	2.8	2.5
Acenaphthylene	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
Acenaphthene	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
Fluorene	< 0.01	1.1	< 0.01	1.3	1.1	1.1	1.6	1.9
Phenanthrene	11.6	3.4	4.9	5.4	5.5	2.2	11.1	9.9
Anthracene	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
Fluoranthene	2.1	2.9	11.2	3.4	1.7	1.2	4.3	3.1
Pyrene	1.8	1.7	7.3	1.9	1.1	< 0.01	3.4	2.5
Benzanthracene	1.2	< 0.01	1.6	< 0.01	1.3	< 0.01	3.9	1.6
Chrysene	1.5	1.2	4	1.2	1	< 0.01	2.8	< 0.01
Benzo(b)fluoranthene	1.7	< 0.01	5.1	1.1	1.6	< 0.01	4.2	1.1
Benzo(k)fluoranthene	< 0.01	< 0.01	1.2	< 0.01	< 0.01	< 0.01	1.6	< 0.01
Benzo(e)pyrene	1.1	< 0.01	1.1	< 0.01	1	< 0.01	1.9	1.4
Benzo(a)pyrene	1.2	< 0.01	2.3	< 0.01	1	< 0.01	2	< 0.01
Dibenzo[a,h]anthracene	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
Indeno1.2.3CD-Pyrene	< 0.01	< 0.01	1.6	< 0.01	< 0.01	< 0.01	1.4	< 0.01
Benzo(g,h,i)perylene	< 0.01	< 0.01	1.9	< 0.01	< 0.01	< 0.01	1.1	< 0.01
Total PAHs	40.8	27.9	47.6	58.2	31.8	9.10	80.0	186

Fig. 2 Total amount of different molecular weight PAHs on the sampling spots in the test plants

Jia et al. 2018). Concentration of Nap was in the range 3.1 (Litér) and 155 µg/kg (Hajmáskér), with relatively high concentrations also in Hárskút (32.7 µg/kg) and Eplény1 (41.9 µg/kg). Nap is generally one of the dominant PAHs in bioaccumulation studies (Waqas et al. 2014; Busso et al. 2018).

Other dominant compound was Phe in concentration varying from 2.2 to 11.6 µg/kg. Phe was found one of the dominant PAHs in vegetables in a Chinese study (Li et al. 2008). Interestingly, detection rate of anthracene was 0, though this PAHs showed high levels in all vegetable samples (including spinach and cabbage) procured from local wholesale markets in Punjab (Pakistan) (Ashraf et al. 2013).

Of 4-ring PAHs, Pyr and the carcinogenic Cry had the highest concentration in Eplény2 (7.3 and 4.0 µg/kg, respectively), followed by Hárskút (3.4 and 2.8 µg/kg).

Relative abundance of five-ring PAHs was rather high in 4 samples (Pécsely, Eplény2, Tihany, Hárskút) showing

peaks in Eplény2 and in Hárskút. Of five-ring PAHs, BkF, BbF, and BaP are considered typical tracers for fossil fuel combustion and are associated with vehicular emissions (Ravindra et al. 2008). These PAHs contributed to 88% and 80% of this group in Eplény2 and in Hárskút, respectively. Six-ring PAHs, namely Ind and BghiP, appeared only in Eplény2 and Hárskút. Their total concentration amounted to 3.5 µg/kg in Eplény2 and 2.5 µg/kg in Hárskút. BghiP is considered a marker of gasoline exhaust emissions (Eiguren-Fernandez et al. 2004). Analysis of individual land-use patterns might give some explanation: Hárskút is situated in a mountainous area and is a Natura 2000 site. However, there is a farm in the neighborhood of the sampling site and high pollution can be explained by the inputs of fossil fuel from farm vehicles which generally pose high environmental load (Ene et al. 2012). Eplény2 is in the proximity of the railway; thus, diesel-powered engines might provide the pollution source.

Fig. 3 Percentage contribution of different molecular weight PAHs on the sampling spots in the test plants

The concentration of total PAHs was the in the range of 9.1 µg/kg dry wt (Nagyvázsony) and 185 µg/kg dry wt (Hajmáskér) (Table 2). Comparing these values with those reported in the literature, Abou-Arab et al. (2014), e.g., measured 8.9 µg/kg total PAH in spinach (*Spinacia oleracea* var.) collected in the urban region of Cairo (Egypt). Similarly, low accumulation was found in spinach in urban gardens of Sao Paulo (Brasil) (Amato-Lourenco et al. 2017). In this study, no PAH accumulation was found in the majority of the gardens; PAH concentration of 7.4 µg/kg was detected only in one garden. Similar values were reported by Ashraf and Salam (2012) in a case study from Saudi Arabia (10.2 µg/kg) and by Ashraf et al. (2013) in a case study from Pakistan (11.5 µg/kg).

However, much higher values were detected in the study of Jia et al. (2018). In this case, the total concentrations of 16 PAHs in spinach collected from near industrial areas of Shanghai ranged from 223.3 to 458.0 µg/kg.

In addition to spinach, Brassicaceae species are also widely used in bioaccumulation studies as they are generally considered good accumulators of PAHs (Franzaring 1997; Xiong et al. 2017; Zhang et al. 2018). Mo et al. (2009) used different *Brassica* species to assess the concentration of 16 PAHs in vegetable samples collected from nine farms of the Pearl River Delta, South China. The concentration of accumulated PAHs were as follows (mean values are given): *Brassica parachinensis* (flowering Chinese cabbage) 438 µg/kg, *Brassica chinensis* (paitsai) 950 µg/kg, and *Brassica juncea* (mustard) 1790 µg/kg. Li et al. (2008) used leafy vegetables including lettuce and Brassicaceae species such as cabbage, mustard, and Chinese flowering cabbage in a case study from Shunde (China). Concentration of accumulated PAHs was in the range of 128–1258 µg/kg in these vegetables altogether (not specified by species). Even much higher values were detected in another Chinese study (Zhang et al. 2004) where the levels of 16 EPA PAHs were determined in vegetable samples from Minjiang River Estuary, China. Vegetable samples included Chinese cabbage. Total PAH concentrations varied from 8600 to 111 000 µg/kg dry wt in vegetable samples (not specified by species).

While these studies sometimes report extremely high values, the data reported by Jánská et al. (2006) make regional comparison possible. In their work, the accumulation of total PAHs ranged between 12.34 and 78.09 µg/kg in cabbage samples collected from Southern Moravia (Czech Republic). Even lower accumulation was reported in a Romanian study (Soceanu et al. 2014), where maximum concentration of total PAHs amounted to 8.216 µg/kg in cabbage and 8.131 µg/kg in spinach collected from rural areas.

Bearing in mind, however, that different vegetables will have different accumulation potential (Franzaring and van der Eerden 2000), comparison of our results with other leafy vegetables has only indicative value. Focusing on studies

where lettuce was used as passive monitor, PAH accumulation in lettuce showed a very wide range. In a pot experiment of Gelman (2014), practically no accumulation was detected in experimental rooftop gardens in Helsinki. On the other hand, in the abovementioned study of Jia et al. (2018), the total concentrations of 16 PAHs in lettuce collected from near industrial areas of Shanghai ranged between 132.0 and 319.2 µg/kg. The highest value in our study (Hajmáskér, 185 µg/kg dry wt) is in fact in this range, indicating significant contamination.

3.2 PAH concentrations in soil samples

Similarly to vegetable samples, concentration of 19 PAHs, including the 16 EPA PAHs, was determined in all soil samples. Detect date of individual PAHs (Table 3) was higher than in lettuce samples: 12 PAHs occurred in all soil samples (Methy-Nap, Phe, Flt, Pyr, BaA, Cry, BbF, BkF, BaP, BeP, Ind, BghiP). Only one PAHs (Acy) had a detection rate of 0.

Table 5 summarizes the concentration of individual PAHs and their total concentration. Figure 4 shows the total amount of different molecular weight PAHs in the sampling spots, while Fig. 5 shows their percentage contribution. Flu (in the range of 4.7–107 µg/kg), BbF (4.2–57.9 µg/kg), and Pyr (3.3–86.2 µg/kg) are the major PAH congeners. Similar pattern was reported by Zheng et al. (2014), who detected the dominance of Nap, Flu, Phe, and BbF in the rural soils of Dongjiang River Basin (China). BbF and Pyr were found to be amongst the most abundant PAHs in the study of Wang et al. (2016).

Concentration of total PAHs was in the range of 31.6 (Litér) and 595.1 µg/kg (Hárskút). Four-ring PAHs showed the highest tendency to accumulate in the majority of samples; their concentration was in the range of 14.9 µg/kg (Litér) and 304.7 µg/kg (Hárskút). Of five ring PAHs, BbF, BkF, BeP, and BaP occurred in all of the samples while the detection rate of DahA was much lower, it occurred only in Hárskút, in concentration of 5.3 µg/kg. Maliszewska-Kordybach et al. (2009) analyzed the spatial distribution of individual PAHs in agricultural soils in over 200 localities in Poland and found that the high molecular weight (HMW) hydrocarbons (groups of 4 + 5 + 6 rings PAHs) represented 72.6% of the total PAH content. Dominance of HMW PAHs in contaminated soils was reported in, e.g., some Chinese studies as well (Wang et al. 2016; Wang et al. 2017). Analysis of dry deposition and soil samples indicated that soil acts as a sink of HMW PAHs throughout the year (Bozlaker et al. 2008). Wet deposition also resulted in the accumulation of HMW PAHs (Zavgorodnyaya et al. 2019).

Comparing the individual villages, Hárskút showed outstandingly high concentrations. Concentration of 3-ring PAHs amounted to 87.1 µg/kg, 4-ring PAHs to 304.7 µg/kg, 5-ring PAHs to 150.6 µg/kg, and 6-ring PAHs to 30.4 µg/kg. Roughly, amount of individual PAHs was one

Table 5 Concentration of PAHs in soil samples. Priority PAHs are given in *italics*

PAH compounds	PAHs concentration ($\mu\text{g/kg}$ dry wt) in soil samples							
	Pécsely/ T	Nagyvázsony /T	Eplény2/ T	Eplény1/ T	Tihany/ T	Litér/ T	Hárskút/ T	Hajmáskér/ T
<i>Naphthalene</i>	13.3	33.1	5.1	154	< 0.01	1.1	17.3	15.7
2-methyl-naphthalene	1.5	2.6	1.5	5	1.9	1.2	3.1	1.5
1-methyl-naphthalene	< 0.01	1.2	< 0.01	3.1	1	< 0.01	1.9	< 0.01
<i>Acenaphthylene</i>	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
<i>Acenaphthene</i>	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	4.8	< 0.01
<i>Fluorene</i>	< 0.01	< 0.01	< 0.01	1.6	1	< 0.01	4	< 0.01
<i>Phenanthrene</i>	5	6	5.4	13.1	4	2.3	56.5	5.8
<i>Anthracene</i>	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	21.8	< 0.01
<i>Fluoranthene</i>	7.4	8.6	10.8	4.7	8.6	6.9	107	8.4
<i>Pyrene</i>	5.5	6	7.2	3.3	5.8	5	86.2	6.1
<i>Benzantracene</i>	1.8	1.6	2.4	4.5	1.4	1.5	68.2	2.1
<i>Chrysene</i>	1.5	1.6	4	4.4	3.3	1.5	43.3	3.4
<i>Benzo(b)fluoranthene</i>	4.3	4.8	5.9	6	4.2	4.3	57.9	4.7
<i>Benzo(k)fluoranthene</i>	1.3	1.5	1.6	2.1	1.3	1.4	24.3	1.1
Benzo(e)pyrene	1.1	1.1	1.6	2.4	< 0.01	1.1	38.7	1.2
Benzo(a)pyrene	2	2	3.3	3.2	1.8	1.5	24.4	2.8
<i>Dibenzo[a,h]anthracene</i>	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01	5.3	< 0.01
<i>Indeno 1.2.3CD-Pyrene</i>	1.3	1.4	2.3	2.2	1.2	1.1	17.7	2
<i>Benzo(g,h,i)perylene</i>	1.2	1.9	2.2	1.7	1.4	2.7	12.7	2.00
Total PAHs	36.9	73.4	53.3	211	36.9	31.6	595	56.8

magnitude higher than in the other samples (Table 5). For rhizosphere soils, Maliszewska-Kordybach (1996) suggested a contamination classification system, defining the following categories: weakly contaminated ($> 200 \mu\text{g/kg}$), contaminated ($600\text{--}1000 \mu\text{g/kg}$), and heavy contamination ($> 1000 \mu\text{g/kg}$). Of our samples, Eplény1 and Hárskút fall into the contaminated category; the others are classified as weakly contaminated. This classification system, however, cannot be fully used in this study as soil PAH concentrations refer only to a

2-month exposure time. However, the 50/2001. (IV. 3.) Decree of the (Hungarian) Government defines a 1 mg/kg limit value for the PAH content of wastewater sludges intended for agricultural use.

3.3 Source appointment

For identifying possible sources of different PAHs, traditionally established PAH isomer ratios were used which

Fig. 4 Total amount of different molecular weight PAHs in soil samples

Fig. 5 Percentage contribution of different molecular weight PAHs in the soil samples

have also been applied to allocate sources of these compounds in vegetables (e.g., Li et al. 2008) and in soils (e.g., Yang et al. 2017b). Fluoranthene to fluoranthene plus pyrene (Flt/Flt+Pyr) ratio > 0.5 indicates grass, wood, or coal combustion as the potential source (Yunker et al. 2002). This value was exceeded in all of the lettuce samples, showing the contribution of household heating and biomass combustion. Burning agricultural waste in gardens is a common practice, providing additional input.

While Flt/(Flt+Pyr) ratio could be calculated for all vegetable samples, the other ratios provided much less information. Benz[a]anthracene to benz[a]anthracene plus chrysene (BaA/BaA+Cry) ratio over 0.35 has been defined to indicate combustion of vegetation and fossil fuel (Yunker et al. 2002). In our study, this ratio could be calculated for only four lettuce samples: Pécsely, Tihany, Eplény2, and Hárskút. In these cases, BaA/(BaA+Cry) ratio was in the range of 0.37 (Eplény2) and 0.5 (Tihany), showing the contribution of pyrogenic sources.

Indeno[1,2,3-cd]pyrene to indeno[1,2,3-cd]pyrene plus benzo[g,h,i]perylene (Ind/Ind+BghiP) ratios between 0.20 and 0.50 likely imply liquid fossil fuel combustion while ratios > 0.50 imply grass, wood, and coal combustion (Yunker et al. 2002). As these 6-ring PAHs occurred only in lettuce samples exposed in Eplény2 and Hárskút sites, ratios could be calculated only for these samples. In Eplény2, this ratio was 0.46, indicating fossil fuel combustion as the potential source, while in Hárskút, it amounted to 0.56, indicating biomass combustion. Considering individual, characteristic PAHs, DbA had a detection rate of 0. This compound is typically associated with coal combustion (Pant et al. 2017).

In the soil samples, similarly to lettuce samples, Flt/(Flt+Pyr) ratio exceeded 0.50, indicating the input from biomass burning, regardless of the size or location of the village.

Ind/Ind+BghiP ratios could be calculated for all of the soil samples: values were between 0.20 and 0.50 in case of Nagyvázsony, Tihany, and Litér (which indicates liquid fossil fuel usage), at the transition point of 0.5 in Hajmáskér and > 0.50 in Eplény1, Eplény2, Hárskút, and Pécsely (which indicates biomass burning, most possibly grass and wood) (Yunker et al. 2002).

BaA/(BaA+Cry) ratio was < 0.35 in case of only 1 village, Tihany (0.30) which implies either petroleum or combustion. In the other sampling sites, it was in the range of 0.38–0.61 which implies combustion of vegetation and fossil fuel (Yunker et al. 2002).

For soil samples, BaA/(BaA+Cry) ratio against Flt/(Flt+Pyr) ratio as well as Ind/Ind+BghiP ratio against Flt/(Flt+Pyr) ratio was plotted (Figs. 6, 7). Plotting BaA/(BaA+Cry) ratio against Flt/(Flt+Pyr) ratio indicates the high input from combustion in general for the majority of the villages.

Fig. 6 Cross-plot of BaA/(BaA+Cry) ratio against Flt/(Flt+Pyr) for soil samples

Fig. 7 Cross-plot of Ind/(Ind+BghiP) ratio against Flt/(Flt+Pyr) for soil samples

The cross plot of Ind/(Ind+BghiP) ratio against Flt/(Flt+Pyr) ratio, however, distinguishes two groups of villages: in case of Eplény1 and 2, Pécsely, Hajmáskér, and Hárskút, the main input is biomass combustion while in case of Litér, Tihany, and Nagyvázsony, petroleum combustion might provide the main source.

Considering the two samples collected in Eplény, it is interesting to note that they are having very different composition, though the two sampling spots are situated approximately 80 m from each other. Eplény1 sampling site was especially chosen to represent roadside conditions: approximate load is 8000 vehicles per day, of which 6500 are light-duty and 1500 are heavy-duty vehicles (<https://internet.kozut.hu/kozerdeku-adatok/orszagos-kozuti-adatbank/forgalomszamlalas/>). In an early study of Larsson and Sahlberg (1981) accumulation of PAHs in lettuce was assessed. Samples were grown at 12- and 50-m distance from a Swedish highway. Significantly higher accumulation was found at the vicinity of the highway, with the concentration of BghiP 0.5 µg/kg vs. 10.8 µg/kg. Dan-Badjo et al. (2007) placed ryegrass (*Lolium perenne*) pots in the vicinity of a highway and found that the high molecular weight PAHs (5 and 6 rings) represented almost 23% of the total PAH concentrations. In a following study, however, spatial distribution of accumulation was depicted (Dan-Badjo et al. 2008). It was recorded that concentration of accumulated PAHs was the highest between 0 and 10 m on both sides of the road than it started to decrease rapidly. However, source appointment did not show extra burden from liquid fossil fuel combustion in Eplény1, though the pots were placed approximately 5 m from the road. In this soil sample, concentration of total PAHs was relatively high (211.3 µg/kg), but prevalence of two-ring PAHs was experienced; concentration of naphthalene was 154 µg/kg. Nap was also a dominant PAHs in the vegetable sample, with the concentration of 41.9 µg/kg.

4 Conclusion

A pot study was conducted using lettuce plants as passive monitors in small- and medium-sized villages. Although concentration of accumulated PAHs in lettuce plants moved in a rather wide range, higher values were comparable to results reported from relatively polluted regions in the world. Source appointment revealed that biomass burning provided important source of PAHs, involving household heating and burning of agricultural waste in the gardens as well. In addition, traffic-related emissions also had high contribution in some of the sites assessed. The fact that both the lettuce and soil samples showed the highest pollution in the Natura 2000 site Hárskút draws our attention to how important it is to analyze individual pollution sources.

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